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CYTOGENETIC STUDIES IN POA PRATENSIS AND ITS HYBRID WITH POA ALPINA

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I. INTRODUCTION.

IN 1933 MÜNTZING published his paper on »Apomictic and sexual seed formation in *Poa*» and was the first to demonstrate apomictic seed formation in *Poa pratensis*. *Poa pratensis* is one of the most important species of grass in our pastures, and therefore breeding work with this species is of the greatest importance. MÜNTZING's findings had far-reaching effects on this work. It was therefore natural that an investigation should be made on the occurrence of apomixis in the breeding material of *Poa pratensis* I had in charge at that time at Weibullsholm. Gradually this material was greatly increased, seeds and plants being collected on journeys or acquired in other ways from Scandinavia, Finland, Germany and England.

The main purpose of the investigations was originally to study the occurrence of apomixis in the breeding material, but owing to the results and experiences gradually obtained they came to embrace several other problems. They cannot yet be regarded as concluded. Up to the present the investigations have been mainly concerned with the following problems in *Poa pratensis*:

1) Gametophyte and sporophyte development in apomictic and sexual forms.

2) The occurrence of aberrants in apomictic forms and their inheritance.

3) The presence of apomixis in material from different habitats.

4) The occurrence of polymorphism and in conjunction therewith the morphology of forms having different chromosome numbers.

5) The origin of apomixis.

6) The origin of hybrids with *Poa alpina* and their progenies.

Some of the results obtained have been published previously (ÅKERBERG, 1936 a and b, 1938, 1939 and 1940). In these papers a special account of methodical questions is given and is therefore not

repeated here. For the same reason no review of other similar investigations in *Poa* is given in this »Introduction».

In the following pages the significance of found values has in many cases been tested. To obtain the P-values corresponding to the found variance quotient, χ^2 and t the tables in »Biologisk variationsanalys» by BONNIER and TEDIN (1940) are used. For the average value the symbol M is used and for the number of variants the symbol n.

During the years 1935—1938 the material was cultivated at Weibullsholm, in 1939 at Svalöf and Lännäs, Undrom, the work being now carried on exclusively at the last-mentioned station. I wish here to express my deep gratitude to Prof. H. NILSSON-EHLE, late head of the Swedish Seed Association, who was also my teacher in genetics, to Prof. Å. ÅKERMAN, the present head of the institute, both of whom have in various ways made it possible for me to accomplish my work, and to Dr. G. NILSSON-LEISSNER, who rendered valuable assistance during the year the material was located at Svalöf.

I also beg to acknowledge my indebtedness to Director H. WEIBULL for the appreciation he showed in my theoretical work during my stay at Weibullsholm's Plant Breeding Institute.

Ever since the beginning of my work I have had the benefit of receiving Prof. A. MÜNTZING's valuable advice and wide experience, not least during the collocation of the material. For that and for granting me facilities to carry on my work at the Institute of Genetics at Lund, I have pleasure in acknowledging my indebtedness and thanks. To Dr. Å. GUSTAFSSON I wish to tender my sincere thanks for his encouragement and advice and for the great interest he has always shown in my material.

I am also indebted to many persons for valuable assistance in various ways, discussions, collecting seeds, etc. I have to mention Prof. J. A. NANNFELDT and Dr. N. HYLANDER, Uppsala, fil. mag. C. L. KIELLANDER, Stockholm, and Mr. Ö. NISSEN, Ås, Norway, colleagues and friends at Svalöf and Lund, and all my co-workers while at Weibullsholm. Thanks to Prof. G. TURESSON, Uppsala, I have received a large material of *Poa pratensis* from North America for cytological analysis. Only part of this material is included in the present investigation. Part of the embryological and cytological investigations were carried out while studying at the Erwin Baur Institute, Müncheberg, and John Innes Horticultural Institution, London, by the kind courtesy of Prof. W. RUDORF and Dr. C. D. DARLINGTON, to whom I wish to tender my sincere thanks.

Finally, I have to mention that the accomplishment of the work was to a great extent rendered possible and facilitated by the valuable and untiring help given me by my wife, Mrs. INGA ÅKERBERG, and Miss MAGNA PALM, Svalöf.

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II. EXPERIMENTAL.

1. INVESTIGATIONS OF 12 SPECIALLY SELECTED STRAINS.

A. TAXONOMY.

The genus *Poa* of the family Gramineae has been subjected to widely different classification. A survey and a critical examination of the different suggestions put forward have been published by NANNFELDT (1935). On the basis of this examination NANNFELDT divides the genus into 10 subgenera, one being submitted as somewhat doubtful. In NANNFELDT's classification *Poa pratensis* L. belongs to the subgenus *Stoloniferae*, to which group *Poa arctica* R. BR. has also been referred.

Poa pratensis L. is very widely distributed in Europe, Asia, North Africa and North America, and is an exceedingly polymorphous species. LINNÉ distinguished the two species *Poa pratensis* L. and *Poa angustifolia* L., both of which will be combined under the specific name of *Poa pratensis* L. (coll.) in the following pages. Since LINNÉ's days a large number of types, occasionally with the inscription of specific names, at other times described as subspecies, varieties, etc., have been distinguished among the Scandinavian form complex, to which variation we shall strictly confine ourselves in this paper. The most extensive classification was made by LINDMAN, who in »Skandinaviens Flora» (HOLMBERG, 1926) describes no less than five species: *Poa pratensis*, *P. angustifolia*, *P. alpigena*, *P. subcoerulea* and *P. irrigata*, in addition to subspecies, varieties and forms. BROWN (1939) made a summary of earlier (not Scandinavian) treatments of the species. The degree to which the species had been segregated varied between 5 and 22 entities. LINDMAN's classification into *species* is no longer generally adopted among taxonomists, most investigators supporting NANNFELDT's (1934) view, who says that »at present it seems, however, as if we had better regard *P. pratensis* as one very polymorphous species, the LINDMAN's segregates being all very complex and connected with each other through too many transitional forms» (cf. HYLANDER in »Vilda växter i

Norden», published by LAGERBERG, 1937; HIITONEN, 1933; NORDHAGEN, 1940, and also NANNFELDT, 1940).

All these authors, however, divide *Poa pratensis* L. into certain form complexes, in which the main types are characterized by certain features in common, but from which there are transitional forms to other complexes. These form complexes thus correspond essentially to LINDMAN's above-mentioned species. There is therefore a satisfactory agreement as to what form complexes should be referred to *Poa pratensis* L. (coll.) (i. e. originally *Poa pratensis* L. and *P. angustifolia* L.). There are only some doubts as to certain mountain forms. Such extreme forms will not, however, be dealt with in this work. The more important form complexes into which practically all systematists now divide the Scandinavian variation of *Poa pratensis* L. (coll.), and which, as will be discussed below, are separate with regard to habitat and distribution, are: 1) *eu-pratensis* HIITONEN, 2) *angustifolia* (L.) LINDB. FIL., 3) *irrigata* (LINDM.) LINDB. FIL. and 4) *alpigena* (FR.) HIITONEN. Sometimes *irrigata* is divided into two main series, comprising shore and wood forms (HYLANDER), occasionally a further form complex, *subcoerulea* (NORDHAGEN), is distinguished, which is closely related to *irrigata*.

A short summary of the chief morphological characteristics of these four form complexes has been kindly given me by HYLANDER, and is given below for the sake of comparison with the strains of *Poa pratensis* L. (coll.) selected for investigation. (In the following pages the species will be written simply *Poa pratensis*.)

1) The form complex *eu-pratensis* HIITONEN. This complex comprises a miscellaneous series of forms, which in their morphological characters are intermediate between the following two complexes, and are very variable in size and tillering. Pure green, not pruinous, leaves of sterile shoots not folded, varying greatly in length and breadth, sheaths not very firm, sterile shoots not firmly closed as in *angustifolia* but frequently many together round the frequently numerous culms, thus forming ± dense and leafy groups, rhizomes (suboles) ± numerous (often very numerous), coarse. Panicle ± pyramidal—broadly egg-shaped, in rather many storeys, lower branches as a rule 3—4 together, fruit panicle not regularly contracted. Spikelet rather large, relatively short, glumes not extended. Fruit smaller than in *irrigata*.

2) The form complex *angustifolia* (L.) LINDB. FIL. Sheaths very firm, culms (often several together) together with firmly closed (intra-vaginal) sterile shoots forming small but very dense tufts, rhizomes few, short and coarse. Leaves very narrow, folded. Not pruinous. Culms often long (about $^1/_2$ m. or more). Panicle with numerous storeys, lowest having frequently 4 branches, relatively narrow (in

anthesis ± egg-shaped), in fruiting stage unilaterally (plume-like) inflexed. Spikelets small, often (even very) faintly purplish, very numerous in dense clusters.

3) The form complex *irrigata* (LINDM.) LINDB. FIL. Culms single or few, surrounded by a few sterile shoots with slender sheaths, not densely tufted. Rhizomes long and slender. Leaves, upper part of culm and spikelet intensely pruinous, with bluish or greyish blue ground colour. Panicle with several storeys, the lowest consisting usually of 2—3 branches, panicle branches (especially lower ones) ramose, somewhat pendent at anthesis, panicle branches also standing out at the fruiting stage. Culms relatively low. Leaves of sterile shoots ± plane (at any rate not folded when healthy), of varying length and breadth. Spikelets large, relatively few, glumes very long and narrow, fruit large.

This form complex comprises two rather distinct series of forms, which in nature appear to be separate both morphologically and ecologically.

a) The shore forms (more strictly seashore forms): short (usually 10—15 cm.), leaves short and wide, panicle ± tinged deeply violet, broadly pyramidal.

b) The forest forms. These include a type identified as v. *nobilis* (LINDM.). This type occurs on woodland paths, in mountain caves, etc., in loose, humous soil, culms as a rule 20—30 cm., solitary, rhizomes very long, thin as threads, leaves (leaves of sterile shoots often one or two decimetres in length, relatively narrow) and panicle silvery white, pruinous, basal panicle branches very long, slender and lax.

4) The form complex *alpigena* (FR.) HIITONEN. Subterranean shoot-system richly branched, forming short rhizomes (extra-vaginal shoots), which are soon bent upwards and terminated above the ground with a sterile rosette; culms solitary, mostly without surrounding sterile shoots. Leaves of sterile shoots plane or (loosely) folded. Not pruinous. Panicle narrow (at least narrowed at the apex). Often with numerous spikes, with many storeys. Spikelets moderately large (rather variable), often tinged purplish violet, glumes and pales brown-tipped and often [especially the pales (lemmata)] sharply pointed, panicle frequently very variegated.

As the main purpose of the present work was to carry out genetical and cytological investigations of the known Scandinavian varieties of *Poa pratensis*, the first task was to find and experimentally examine good types belonging to each of these four complexes. Altogether 12 strains were selected and analysed, and were given the following designations: Ä 787, 701, 702, 703, 704, 5303, 746, G 258, 768, G 125, 709 and 813. These 12 strains have in some way or other formed part of previously published investigations. These investigations showed that 10 of the 12 strains were mainly apomictic, while the remaining two, 709 and 813, appeared to be highly sexual. Apomixis or sexuality was established by investigations of progenies and chromosome determinations, in some cases also by embryological studies. In the determination of the characteristics of the known apomictic species, not only the original mother plants were employed but also to a great extent the indubitable maternal progeny plants. Thus of the said 12 strains there

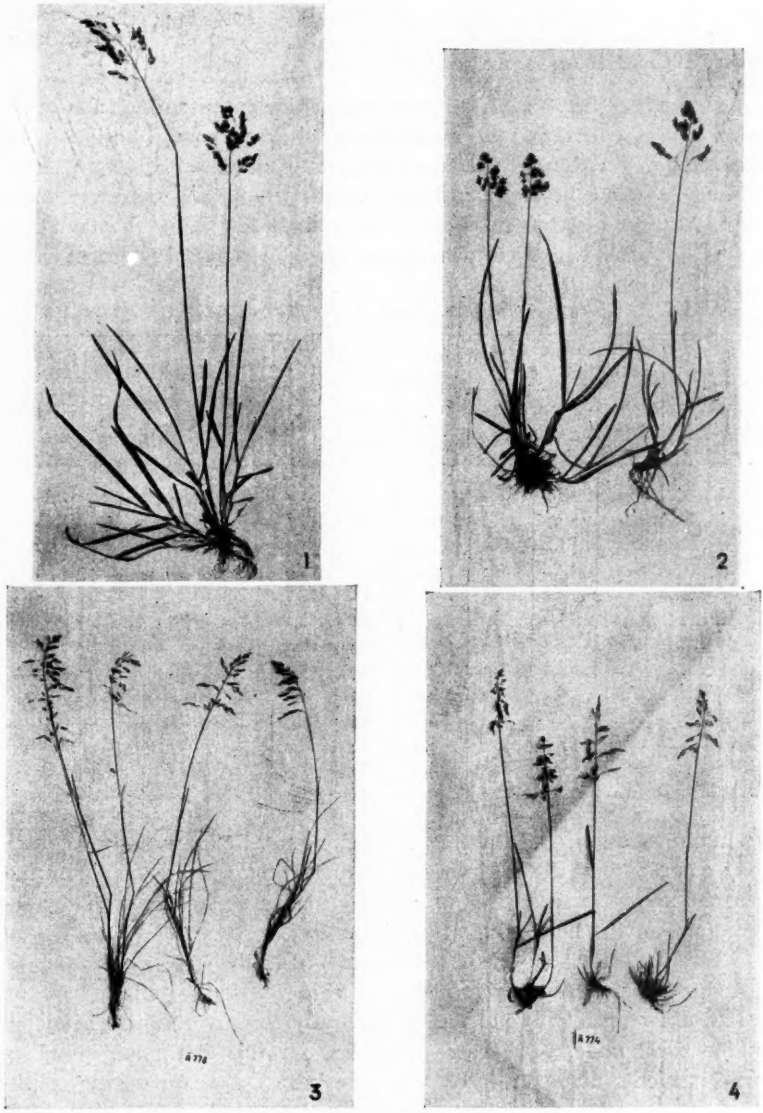
existed a very extensive experimentally produced material, and in a later chapter an attempt will be made to supplement and verify the results obtained by material collected from nature.

B. THE MORPHOLOGY OF THE 12 STRAINS.

The morphological description is based on plants grown in culture. An important question in this connexion is the extent to which the different morphological characters are influenced by environmental factors. No definitive answer can yet be given with respect to the characteristic properties of the form complex. But many of the findings from the investigated material point to the desirability of a thorough experimental investigation of the influence of the environment on these properties. This should not be taken to imply that typical *irrigata* and typical *angustifolia* collected in nature were not obtained in a typical form in the cultures. That was most frequently the case. Nevertheless it appears as if certain properties may be modified, and the degree of this modification should be investigated.

As in the case of the material studied in nature, the material raised in culture also exhibited transitional types between, for instance, *angustifolia* and *eu-pratensis* or between *eu-pratensis* and *irrigata*. Of the *alpigena* complex, which has a northern distribution and which is not quite comparable with the others with regard to origin and ecology, the number of types in culture was not so large. Some of them did not develop quite satisfactorily when grown in Skåne. But we nevertheless succeeded in obtaining typical, well-developed representatives of this form complex.

Some of the 12 strains have already been described with respect to their general morphological characteristics (ÅKERBERG, 1936 a). Since then more properties have been measured, therefore it appears to us that a description of all the strains is justified also here. Such measurements were made on all strains with the exception of Ä 787. As all measurements were not made in the same year they had to be referable to a standard, strain 704, so as to be able to compare all strains (Table 1). With the exception of strains 709 and 813, all measurements were made the year following that on which the seedlings were planted out, i. e. the first year in which the plants developed panicles. Strains 709 and 813 were not measured until the third year after planting. Data for height and circumference of the tufts for these two strains, which were sexual, originate from plants obtained after isolation. They are therefore not quite comparable with the values for the other strains. The circumference of the tufts was measured for all strains in the same year, it cannot therefore be associated with the other properties. The measurements of the panicles and leaves are based on at least three culms or leaves per plant respectively. In the case of strains 709 and 813 only one plant was measured. As



Figs. 1—4. — 1. Strain 5303. *Poa pratensis* f. *eu-pratensis*. — 2. 709. *P. pratensis* f. *irrigata*. — 3. Ä 787. *P. pratensis* f. *angustifolia*. — 4. G 258. *P. pratensis* f. *alpigena*.

regards the standard strain 704, at least three plants were measured each year. The data for the measurements of various morphological characters published later on were obtained in the manner described above.

Strain 787 (Fig. 1), the only one for which no measurements were made, agrees entirely in its characters with the above description of *angustifolia*. In the field it was difficult to differentiate strains 701, 702 and 703, and the measurements, too, show only slight divergencies. Strain 701 seems to have a shorter leaf and panicle than the other two strains, and, as will be shown later, has a considerably lower seed

TABLE 1. *Showing morphological properties of the 12 strains.*

Strain	Height in cm.	Panicle length cm.	No. branch lowest node	Apical culm leaf		Penultimate culm leaf		Sterile shoot		Tuft circumference[1] cm.
				lgth cm.	bdth mm.	lgth cm.	bdth mm.	lgth cm.	bdth mm.	
704	86,7	16,1	5,0	6,8	5,7	11,8	6,4	28,4	3,9	140,3
701	83,9	10,1	3,9	7,1	4,1	10,7	4,3	29,7	2,6	136,7
702	82,6	11,7	3,9	7,8	3,9	11,2	4,7	31,6	2,5	139,1
703	83,3	12,3	3,9	8,1	3,9	11,2	4,7	30,1	2,9	137,5
5303	77,0	9,7	3,1	4,2	4,3	7,9	5,5	30,3	4,3	145,1
746	47,0	7,6	3,9	3,2	3,3	6,3	4,5	20,9	3,8	94,2
704	94,0	13,9	4,8	4,7	3,8	8,9	5,2	30,7	3,9	140,3
709[1].....	(49,0)	5,5	2,3	2,1	2,7	3,9	3,2	15,4	3,7	(87,0)
813[1].....	(62,0)	6,0	2,0	2,6	3,5	4,0	3,7	21,1	4,2	(107,0)
G 258 ...	54,0	7,8	5,7	3,1	2,9	3,8	3,0	21,2	4,1	88,3
704	65,0	13,2	5,1	8,3	4,6	11,8	5,8	24,7	4,3	140,3
768	50,0	9,0	5,0	4,5	3,3	6,8	4,7	11,8	4,0	125,7
G 125 ...	46,0	7,7	4,0	4,5	4,0	6,7	4,3	23,0	5,0	105,0

setting in isolation. All three strains belong most closely to the *angustifolia* complex, but differ from typical *angustifolia* in having somewhat broader leaves, good spreading capacity and large spikelets. They are therefore on the borderline of *eu-pratensis*.

Strains 704, 5303 (Fig. 3), 746 and 768 all belong to *eu-pratensis*, but showed a great inter-variation. Strain 704 is the tallest, its culm leaves are long and broad and it generally has five panicle branches at the basal node. The values recorded for this strain are based on measurements made in three successive years, but for plants of the same age each year. The characters measured show very great variation, especially height and the length and breadth of the apical

[1] See text.

leaf. Strain 5303 is somewhat shorter than strain 704, its culm leaves are a little shorter and narrower, whereas the sterile shoots and spreading capacity are as big as those of strain 704. Strain 746 is in all respects vegetatively weaker than strains 704 and 5303, it has shorter and narrower leaves and inferior spreading. The same thing is also true of strain 768, which nevertheless forms a larger tuft. Strain 768 seems to have strikingly short vegetative leaves.

Strain G 125 was obtained as being closely related to *irrigata*, most proximate to a forest form of *irrigata*. In culture it was found to resemble the *irrigata* complex in some respects, in others it agreed more with *eu-pratensis*. It may be regarded as an intermediate form between *irrigata* and *eu-pratensis*. Strains 709 and 813 are typical *irrigata* (Fig. 2). Judging from the properties measured, this relationship can be seen in the low height, the short panicle and the small number of spikelets branches on the basal panicle node. Both strains are intensely pruinous.

Finally, strain G 258 (Fig. 4), which was obtained under the name of *alpigena*, seems to agree well with the description of this form complex. The shape of the panicle and the formation of extra-vaginal shoots in particular were very typical. A striking feature is the large number of panicle branches at the basal node. Judging from the measurements, the strain appears to be vegetatively weak, but this may be due to the change in environment in the south of Sweden being too great.

C. SOMATIC CHROMOSOME NUMBERS.

All fixations of root-tips for somatic chromosome counts were made in MÜNTZING's (1933 a) modification of Navashin's solution. As far as possible two or three plates of each plant were counted, but in some cases only one plate was available for a reliable count. Most of the chromosome numbers for strains 701—704 and the number for strain 709 recorded in the table have been published previously (ÅKERBERG, 1936 a).

The chromosome numbers of all the 12 strains except strain 813 have been determined. As will be shown later, the chromosome number of strain 813 is probably between 90 and 95. The chromosome numbers of several of the strains were determined more than once. Table 2 is intended to give a survey of the chromosome numbers obtained. As already pointed out by MÜNTZING (1933 a) and RANCKEN (1934), it is exceedingly difficult to determine the chromosome number in *Poa*

pratensis, partly owing to the large number of chromosomes, partly because they often lie crowded in the plates. On that account the agreement in the determinations of the chromosome numbers of the different strains must be regarded as satisfactory. In only two strains is the variation in relation to M greater than ± 1 (in strain 704 M = 53, max. 55, min. 52; in strain 768 M = 86, max. 87, min. 84).

The chromosome numbers of the strains vary between 50 and 90.

TABLE 2. *The somatic chromosome numbers of the 12 strains.*

Strain	No. of plants examined	M ±	2n values obtained ±	Form complex
Ä 787	1	50		*angustifolia*
			n = 2 2 3	
701	7	50	49 50 51	*angustifolia—eu-pratensis*
			n = 1 12 2	
702	15	50	49 50 51	» »
			n = 3 3	
703.....	6	50	50 51	» »
			n = 1 3 2	
704	6	53	52 53 55	*eu-pratensis*
			n = 1 1	
5303	2	78	77 78	»
746	1	80		»
			n = 1 2 1	
768	4	86	84 86 87	»
G 125	1	87		*eu-pratensis—irrigata*
709	1	90		*irrigata*
813		90—95[1]		»
			n = 1 1	
G 258	2	83	82 84	*alpigena*

Somewhat lower as well as occasionally higher values have been found in *Poa pratensis* (see p. 87), but the strains cover the most important and the most common chromosome variation in the Scandinavian types of the species. Four of the strains have 2n = 50. One of these strains was evidently *angustifolia*, the other three were on the borderline of *eu-pratensis*. The strains belonging to *eu-pratensis* have 2n = 53, 78, 80 and 86 respectively, while strain G 125, which comes close to *irrigata*, has 2n = 87. The two *irrigata* strains 709 and 813 have a maximum chromosome number of 90 and 90—95 respectively.

[1] See text.

The chromosome number of the *alpigena* strain G 258 is 2n = 83. The three strains 701, 702 and 703, between which no great morphological differences were discernible, have thus the same somatic chromosome number, viz. 2n = 50.

D. DEVELOPMENT OF THE MALE GAMETOPHYTE.

Some data concerning the reduction division in the P.M.C:s of *Poa pratensis* are already published in the literature (MÜNTZING, 1933 a, 1940; RANCKEN, 1934; FLOVIK, 1938, and ARMSTRONG, 1937). In the reduction division in an apomictic strain with 2n = 49 MÜNTZING observed univalents and probably also multivalents. Thus the irregularities were of such a nature that they must lead to gametes with varying chromosome numbers. In the 1940 paper MÜNTZING mentions the reduction division in a sexual strain having 2n = 36, probably arisen by haploparthenogenesis from a strain with 2n = ± 72. This strain is therefore an aberrant, and its reduction division will be dealt with later on. RANCKEN (1934) investigated the reduction division in a normal strain with 2n = 66—67 and found, as in MÜNTZING's strain with 2n = 49, the formation of univalents and multivalents. In the first and second divisions chromosomes were observed scattered irregularly outside the plates and even in the tetrad stage chromosomes were found outside the nuclei. They were assumed to be resorbed in the plasm. Otherwise no divergencies, such as the formation of pentads, hexads, etc., were observed. FLOVIK found irregularities similar to those mentioned in the reduction division in three strains of *alpigena* having 2n = 46, 77 and 84 respectively. But in these strains pentads, hexads, etc., in addition to tetrads, were formed, and even dyads were met with. ARMSTRONG investigated the frequency of cells with univalents in five different strains and found a variation in this respect of from 2,3 to 93 per cent, the average number of univalents per cell being 0,02 and 3,9 respectively. ARMSTRONG very seldom observed micronuclei in the tetrad and later stages. ANDERSEN (1927) also mentions a normal formation of pollen-tetrads.

Our investigations of the reduction division in the P.M.C:s of the strains under discussion have not been particularly extensive. They are limited to the three strains 703, 746 and 709, and were made in sections intended for embryological studies. Moreover, it was found that in stages suitable for studying the E.M.C:s in strain 746 the pollen was as a rule fully developed, and for that reason the material of this strain was very scanty.

If transverse sections are made at an early stage through the pollen-sac, 5—6 P.M.C:s will be found lying together, each forming a sector of a circle. The circle is surrounded by a ring of elongated cells. This is gradually pushed towards the epidermis, while at the same time a cavity is formed in the centre of the circle. The P.M.C:s are thus forced from the centre of the circle, acquire another shape, becoming oval, and finally form a ring of cells, which remains closed until the divisions.

In strain 709 it was possible to study the occurrence of univalents in an occasional flower during first and second divisions. They have been counted at first metaphase as well as at second anaphase, and they seem to occur in rather large numbers. At first metaphase up to 10 univalents have been observed while at second anaphase the number of lagging chromosomes varied between 3 and 8. At least some of these lagging chromosomes never enter the tetrad nuclei. Apart from the occurrence of univalents no irregularities were observed in the formation of the pollen tetrads. No cell-groups having more than four cells were recorded. Reduction division and tetrad formation appear to take place in the same manner in strain 703 as in strain 709. Several univalents were observed also in strain 703, at first metaphase varying between 4 and 11, but otherwise no irregularities occurred in the formation of dyads and tetrads. As already mentioned, the observations in strain 746 are still fewer. It was only possible to study the occurrence of univalents at first metaphase, the number of which was 2—3. Summarizingly, it may, however, be said that pollen formation, i. e. from meiosis in the P.M.C. to complete tetrads, seems to take place in exactly the same way in the sexual strain 709 as in the apomictic strains 701 and 746 and in accordance with that previously described in *Poa pratensis* by MÜNTZING (1933 a, 1940) and RANCKEN (1934).

In earlier papers (ÅKERBERG, 1936 a, 1939) it has been shown that apomictic strains of *Poa pratensis* are pseudogamous, i. e. require pollination for the development of the ovule into seed. But it has also been demonstrated (FR. NILSSON, 1933, 1937; ÅKERBERG, 1936 a) that the seed setting of the apomictic strains is rather variable both after self-pollination and after free flowering. In several cases this variation was found to be associated with the supply of pollen and the quality of the pollen. Haplontic and probably also diplontic sterility (according to MÜNTZING's classification, 1930 a) were observed in the material investigated.

The quantitative and qualitative pollen formation in the 12 strains

has been controlled by studies of the morphology of the stamens and of the pollen. Attempts to investigate the germinability of the pollen in nutrient solution or on the stigmata have failed so far, probably owing to poor technique. In 1937 ARMSTRONG, however, described a method which he had successfully employed in studying pollen germination and the pollen-tube growth in the stigmata. We have not yet been able to test this method.

With regard to strain 704 it has previously been stated that the flowers contain a certain percentage of dried stamens. (When other stamens are ready to dehisce, these defective stamens are small, yellow in colour and as thin as fibres, and are often very difficult to detect.) Later investigations have confirmed this observation (v. Table 3). Of

TABLE 3. *Stamen development in the 12 strains.*

	Ä 787	701	702	703	704	5303	746	G 258	768	G 125	709	813
	Percentage well-developed stamens											
1937.....		100	100	100	61	86	100				not	
1938.....	100		97	100		67	100	100	100	100	investigated	
	Percentage entirely dehisced stamens											
1938.....	100		65	82			90			100		

the other 12 strains only 5303 seems to contain such dried stamens. In order to illustrate further the occurrence in *Poa pratensis* of such strains with dried stamens we may mention an investigation of 30 strains not included in the material treated below. Of these 30 strains 20 had fully developed stamens, 8 showed a frequency of 1—9 per cent dried stamens, while only 2 strains had a frequency of more than 10 per cent, viz. 25 and 34 per cent respectively.

The results of pollen investigations of the 12 strains, both those published in an earlier paper (ÅKERBERG, 1936 a) and those obtained later, are presented in Table 4. The values given are the averages for one to several plants and are based on counts of at least 200 pollen grains per plant, the pollen as a rule being obtained from pollen set free by the stamens. If this procedure failed, the pollen was removed from the stamens. (By well-developed pollen grains is meant morphologically quite faultless pollen.) A certain variation is noticed in the individual strains from year to year, which is of course to be expected as the pollen quality of at least certain strains has varied between different plants of one and the same strain in a year. For instance, in 1938 six different plants of strain 702 had a pollen quality varying between 48

and 94 ($M = 75{,}3 \pm 7{,}3$). Strain 704 was considerably less variable in the same year, the variability, also for 6 plants, ranging between 93 and 99 ($M = 96{,}3 \pm 0{,}9$). The average pollen quality of six of the strains has been calculated for the years 1935, 1937 and 1938. An analysis of the variation in this material goes to show that there exists a probable seasonal difference with respect to pollen quality ($0{,}05 > P > 0{,}01$; P here, and in the following pages if not otherwise stated, the probability for chance variation). In 1935, in which the average pollen quality was the poorest, pollen samples were taken from greenhouse plants, while in 1937 and 1938 they were taken from plants in the open field.

TABLE 4. *Pollen quality and pollen size in the 12 strains.*

Percentage well-developed pollen

Year and habitat	Ä 787	701	702	703	704	5303	746	G 258	768	G 125	M n = 6	709	813
1934, Weibullsholm					95				82			94	95
1935, »		68	74	72	89	85	89				79,5	84	
1936, »				76	98	96	94	95	91	98		79	97
1937, »		72	75	77	96	92	88		88		83,3	80	
1938, »	91	78	75	91	96	85[1]	94	99	95	98	86,2	(94)	
1939, Svalöf		80				80	95—100						
Average 1935, 1937, 1938		72,7	74,7	80	93,7	87,3	90,3				83,1		

Pollen diameter μ

	Ä 787	701	702	703	704	5303	746	G 258	768	G 125	Aver. n = 5	709	813
1937.....		30,8	30,4	30,7			34,2		33,0		31,8	32,9	
1938.....	25,5	27,1	25,9	22,2	32,9	35,0	31,4	34,9	31,3	31,4	27,6		
Average		29,0	28,2	26,5			32,8		32,2		29,7		

The difference in pollen quality between the strains is more evident ($P < 0{,}001$). A difference between two strains of 7,35 lies at the limit of probability of $P = 0{,}05$.

The reason why the stamens in strains 702 and 703 and also, though not to the same extent, in strain 746 do not dehisce is not easy to explain at the present stage. There may be at least two such reasons. In the first place we may have some kind of diplontic sterility which causes a disturbance in the stamen and pollen development and in that way is also the primary cause of the deteriorated pollen quality. But it is also conceivable that the primary cause in some of the strains

[1] In addition there was a plant with 29 per cent well-developed pollen. This plant had throughout defective stamen formation.

is to be found in the haplontic sterility. In the reduction division in strain 703 a very high frequency of univalents was observed, and this should be expected to lead to gametes with different chromosome numbers. In relatively low somatic numbers, as for instance in strain 703 ($2n = 50$), it is possible that some gametes with greatly divergent chromosome sets (combinations) will not be viable. In that way we should obtain a falling off in the pollen quality and possible also a reaction on the stamen development. It is quite conceivable that in the said strain we have a combination of diplontic and haplontic sterility, causing the impaired stamen and pollen development (see further p. 108).

As a further example of the variation in the pollen quality due to environment we may mention that FR. NILSSON (1937) investigated this property in strains 701 and 709 cultivated at Lännäs in the Province of Ångermanland, and obtained 40 and 57 per cent normal pollen respectively, thus considerably lower values than those recorded in Table 4. The value for strain 709 in 1938 is given in brackets, the percentage of well-developed pollen being determined that year in sections from flowers and at a very early stage after the tetrad formation. The value is therefore not exactly comparable with those recorded for other strains, but it does not deviate from the variation of the strain. Thus strains 701—703, all of which had $2n = 50$, have the lowest pollen fertility, but it should be pointed out, however, that strain 704 with a 2n as low as 53 has satisfactory pollen. So far only one value is available for the pollen of strain Ä 787 (91 per cent), therefore nothing definite can be said of this strain.

Even if no results of pollen germination experiments can be submitted, castration and pollination experiments carried out nevertheless point to the probability of certain strains being characterized by low pollen germination. This is true of strains 701 and 702 (ÅKERBERG, 1936 a). It is therefore evident that there are great differences between the quantitative and qualitative pollen formation of different strains but also that both quantity and quality are greatly influenced by modificatory factors.

Table 4 also gives data of the pollen diameter in some of the 12 strains. MÜNTZING has already published (1940) results of pollen measurements in *Poa pratensis*, in which he found an evident correlation between chromosome number and pollen size. In our material, too, there is a correlation in both experimental years. In strains Ä 787, 701—704 with $2n < 60$ the average pollen size in 1938 is 26,7 μ and in

the other strains with $2n > 70$ it is 32,8 μ. It should be noted, however, that strain 704 with $2n = 53$ has a pollen size of 32,9 μ, while that of G 125 with $2n = 87$ is only 31,4 μ. Five strains were investigated in 1937 and in 1938. In 1937 the average pollen size was 31,8 μ, but in 1938 it was only 27,6 (Diff. $4{,}2 \pm 1{,}16$).

E. DEVELOPMENT OF THE FEMALE GAMETOPHYTE.

Only three of the strains, 703, 709 and 746, have been investigated embryologically. Some results of investigations of strains 709 and 746 have already been published (ÅKERBERG, 1939). The fixations were made in the same fixative as that employed for root-tips (p. 9), but with prefixation in Carnoy. At first all slides were stained with gentian violet, but on the recommendation of KIELLANDER safranine, »lichtgrün» and gentian violet are at present employed with good results. Certain difficulties, however, were encountered in section-cutting flowers at a somewhat later stage of development. The pales are very hard and the knife very quickly becomes blunt, the sections therefore get torn and are lost. Attempts will now be made to pre-treat the flowers in hydrofluoric acid.

In our embryological investigations some stages are still missing. But those already observed, however, seem to furnish a satisfactory explanation of the embryological development in the three strains above mentioned. If our results are combined with those obtained by other investigators it should be possible to present a probable picture of the development of the female gametophyte in its main features.

Reference has already been made to ANDERSEN's work (1927) on the embryology of *Poa pratensis*. Her work was thus published before we had any knowledge of the occurrence of apomixis in the species. ANDERSEN found a macrospore development resulting in the formation of 4 spores. She does not, however, mention the nuclear division preceding the formation of the macrospores. The embryo-sac formation was normal, usually from the chalazal macrospore, and occasionally two embryo-sacs were observed in the same nucellus. They were assumed to originate from different megaspores.

ARMSTRONG (1937) ascertains the occurrence of reduction division in the E.M.C:s and assumes that functioning embryo-sacs arise from macrospores, usually here, too, from the chalazal spore. ARMSTRONG also assumes that two megaspores from the same E.M.C., or possibly from two E.M.C:s, can form embryo-sacs in the same nucellus. Further, ARMSTRONG puts forward a theory of sporophyte formation which

attempts, on the assumption of gamete selection (heterogamy) and fertilization, to explain the morphological and chromosomal constancy in the progeny. In an earlier paper (ÅKERBERG, 1939) it was shown that this theory seems to be absolutely untenable, therefore there is no reason to discuss it once more. In that paper it was also pointed out that it was not inconceivable that the material investigated by ARMSTRONG was apomictic with aposporous gametophyte formation, and below we shall put forward further evidence in support of that assumption.

Another investigation of the female gametophyte formation in *Poa pratensis* collected in America was published quite recently (TINNEY, 1940), and the results obtained seem to agree completely with our own results from apomictic strains. TINNEY found:

1) The E.M.C. undergoes meiosis, the formation of univalents and multivalents being ascertained, usually only three megaspores being formed, the micropylar dyad cell often not dividing. In all the observed cases the megaspores degenerated.

2) The functioning embryo-sac develops without meiosis from a vegetative nucellar cell near the chalazal end of the E.M.C.

3) The finished embryo-sac consists of an egg-cell, two synergids, a primary endosperm cell with two nuclei and three antipodal cells.

4) Two embryo-sacs were obtained in the same nucellus, one, owing to a later development or some other cause, being weaker than the other. An occasional purely atypical embryo-sac was met with.

We shall return later to TINNEY's investigations of the sporophyte formation. Here it will only be mentioned that TINNEY never observed nucellar embryony.

In an investigation of a strain of f. compl. *alpigena* KIELLANDER (1941 a) obtained results that were also in agreement with ours and consequently with TINNEY's. In this strain, too, the legitimate E.M.C. does not seem to be of any importance in the formation of a functioning embryo-sac, which arises instead from aposporous E.S. initials. The E.M.C. may degenerate already before the first division, but this generally takes place after the division. The divisions usually result in the formation of only three macrospores, the micropylar dyad cell failing to divide or showing a very abnormal division.

The aposporous E.S. initials appear both at the chalazal and the micropylar end of the nucellus, the number amounting on the average to 2—3. They divide mitotically into a fully developed embryo-sac.

Several E.S. initials may start developing, but owing to competition only one of them usually attains full organisation.

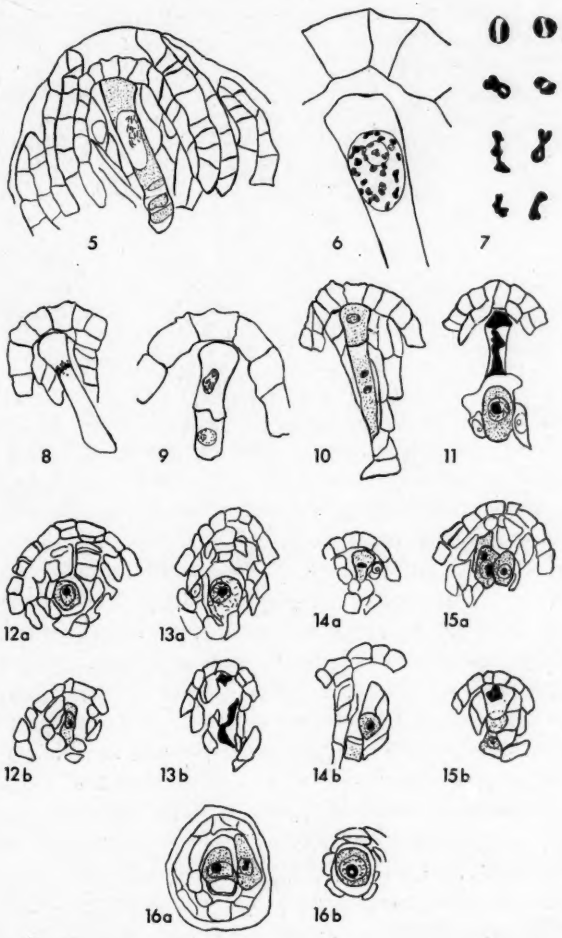
Of the three own strains investigated we shall first deal with the sexual strain 709.

Strain 709 (Figs. 5—11). — Stages from the E.M.C. in the resting stage up to finished megaspores were studied, originating from 32 different flowers. In most of these flowers the E.M.C. was found to be in the resting stage (6) or at meiotic prophase (18). In other flowers we obtained 1 first anaphase, 3 dyads, 1 second anaphase and 2 triads.

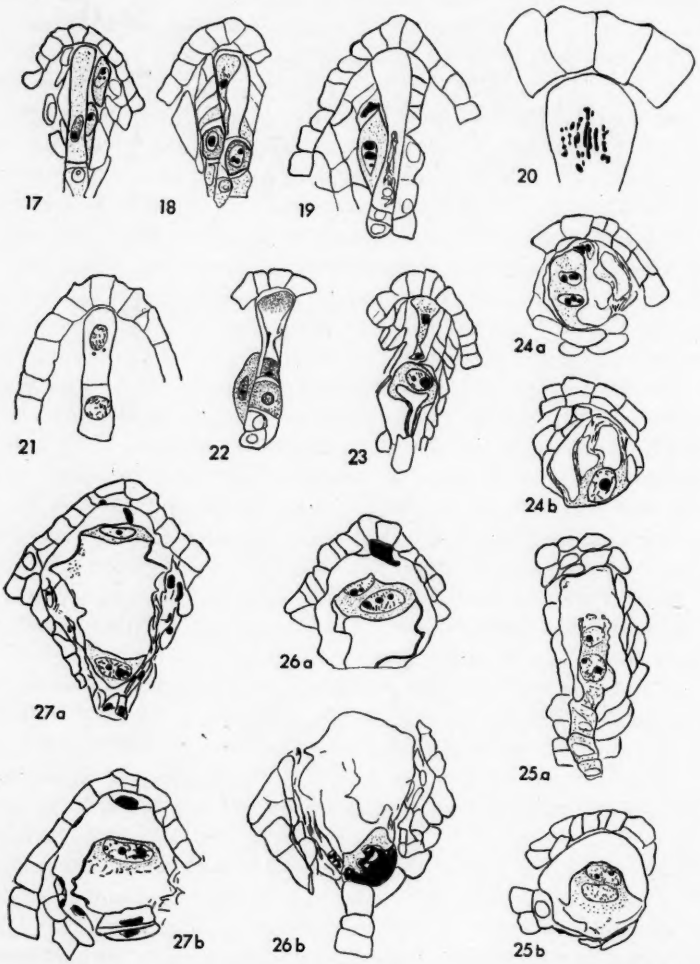
The E.M.C. has a very characteristic appearance and is therefore always easily recognisable. Fig. 5 gives a general idea of the appearance of the E.M.C., its location in the nucellus, etc. Its development up to first metaphase does not seem to differ essentially from a normal meiosis, apart from the occurrence of multivalents and univalents. In one of the diakinetic stages some chromosome »pairs» could be studied, see Figs. 6 and 7. Most of them were interpreted as bivalents, in a few solitary cases at any rate trivalents were observed. At metaphase (Fig. 8) at least three univalents occurred, and at first anaphase three univalents, which had divided, were seen. In the dyad stage the nuclei of the dyad cells seem to be in different stages of development (Fig. 9), and later stages indicate that only one of the dyad cells divide, in accordance with the finding of both TINNEY (1940) and KIELLANDER (l. c.). [In an earlier paper (ÅKERBERG, 1939) it was stated that 4 megaspores were formed in strains 709 and 746. A re-investigation of the material, however, points to the probability of only 3 megaspores being formed in strain 709 and possibly also in strain 746 (see below).] In the observed cases of strain 709 it is the chalazal dyad cell that divides (v. Fig. 10) and the lowest cell of the triad that develops into the E.S. (v. Fig. 11).

In none of the flowers investigated was any cell observed that could be assumed to constitute or to develop into an aposporous E.S. initial. Such aposporous E.S. initials may occur, since, as will be shown below, polyembryony and the development of embryos from unreduced male gametes may take place in strain 709.

Strain 746 (Figs. 17—29). — About the same stages of development of the E.M.C. were observed in this strain as in strain 709. In strain 746, however, a few cases with the E.S. in a more or less finished organisation were met with. Altogether 55 flowers were studied, and the distribution of the different stages of development of the E.M.C. is shown in Table 5. The table also gives the frequency of aposporous



Figs. 5—16. The development of the female gametophyte in 709 and 703. — Figs. 5—11. 709. — 5. The integuments and the nucellus with an E.M.C. — 6. Diakinesis, all configurations not drawn. — 7. Bivalents (one trivalent?) from 6. — 8. Metaphase. — 9. Interkinesis. — 10. Second anaphase. The dyad cell at the micropylar end not dividing. — 11. The three megaspores, two of them degenerating. — Figs. 12—16. 703. (*a* and *b* adjacent sections from the same nucellus). — 12 *a*. An aposporous E.S. initial. — *b*. Degenerating? E.M.C. — 13 *a* = 12 *a*. — *b*. Degenerating dyad. — 14 *a*. E.M.C. at metaphase. — *b*. = 12 *a*. — 15 *a*. Two aposporous E.S. initials and the developing megaspore. — *b*. The three megaspores, two of them degenerating. — 16 *a*. To the left an aposporous E.S. initial, to the right a megaspore. — *b*. The same aposporous E.S. initial as in *a*.



Figs. 17—27. The development of the female gametophyte in 746 (*a* and *b* adjacent sections from the same nucellus). — 17. E.M.C.. and two aposporous E.S. initials. — 18. Degenerating E.M.C. and two aposporous E.S. initials. — 19. An aposporous E.S. initial encroaching upon the E.M.C. (in prophase). — 20. Metaphase in E.M.C. — 21. Interkinesis in E.M.C. with two lagging chromosomes. — 22. Probably three megaspores and an aposporous E.S. initial. — 23. An aposporous E.S. initial coming from the chalazal region and two degenerating megaspores, the third in another section. — 24 *a*. A legitimate E.S. with two nuclei and two degenerating megaspores, pushed aside from an aposporous E.S. initial, seen in *b*. — 25 *a*. A

TABLE 5. *Showing occurrence of aposporous E.S. initials in strain 746.*

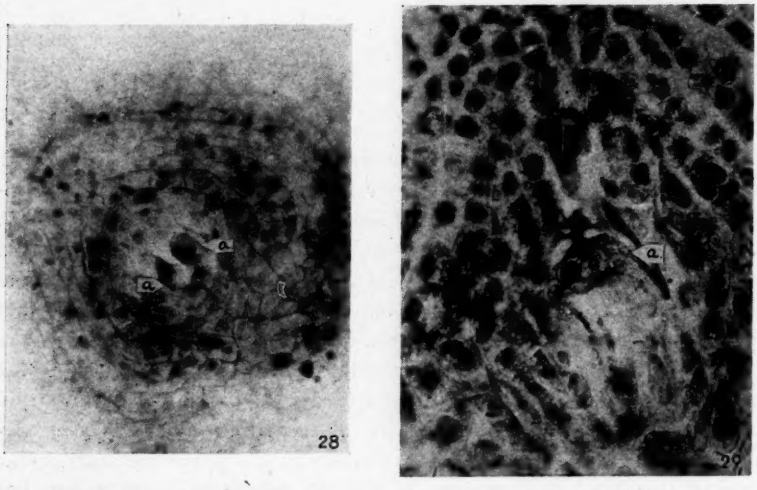
E.M.C. stages	Number of obs. stages	Number of cases with	
		1 aposporous E.S. initial	2 aposporous E.S. initials
Resting stage	22	11	3
Prophase	16	9	2
Leptotene	1	1	—
Pachytene	5	3	—
Diakinesis	1	1	—
Metaphase	1	—	—
Interkinesis	1	—	—
Tetrad?	2	2	—
E.S.	6	6	—

E.S. initials, of which altogether 38 were recorded. It can be assumed, however, that this number is rather low, for the aposporous E.S. initials are not always observed in the same section as the normal E.M.C. It was mentioned above that sections may have been lost, and on that account it is quite conceivable that adjacent sections of the normal E.M.C. were occasionally destroyed. This would therefore imply that certain aposporous E.S. initials were not observed.

In the majority of cases the development of the E.M.C:s up to the metaphase stage seems to follow practically the same lines as in strain 709 described above. Occasionally, however, an incipient degeneration of the E.M.C. was observed already in the prophase stage. One case of this kind is illustrated in Fig. 18, where the protoplasm has contracted and at the lower end consists of only a thin thread. Univalents were observed both at metaphase and at interkinesis, in the latter stage as lagging chromosomes. Both tetrad stages were somewhat difficult to interpret. In one of them (Fig. 22) there seems in all probability to have been only three megaspores, i. e. in this case, too, one of the dyad cells has not divided. Another case indicating the probable occurrence of triads is illustrated in Fig. 24 *a*, the legitimate E.S. having reached the binucleate stage. The appearance of the E.S. and its location in the nucellus are characterized here by the occurrence of aposporous E.S. initials. Another example of a legitimate E.S. in a binucleate stage is shown in Fig. 25 *a*, in which the E.S., however,

degenerating legitimate E.S. with two nuclei. — *b*. An aposporous E.S. with two nuclei. — 26 *a* and *b*. An aposporous E.S. with two nuclei and rests of the megaspores at micropyle and chalaza. — 27 *a* and *b*. An aposporous ? E.S. with four nuclei; the fourth in another section.

appears to be in process of degeneration. Whether this is a result of the aposporous E.S. initial or not is difficult to say. That can hardly be the sole reason. The degeneration of the legitimate E.S. very probably takes place very rapidly, even when it has reached the binucleate stage. In the later stages of the development of the aposporous E.S. only small rests of the products of the E.M.C. (megaspores, 1—2 nucleate E.S.) were observed. Thus no inference can be drawn as to the origin of the E.S. simply by studying its later stages of development.



Figs. 28—29. Microphotos from 746. — 28. Two aposporous E.S. initials (*a*). E.M.C. between them in another section. — 29. = Fig. 23. An aposporous E.S. initial (*a*), the dark shadows above it the degenerating megaspores. — (Photo: 28. H. C. OSTERSTOCK; 29. O. MATTSSON.)

As a rule the aposporous E.S. initials appear at the chalazal end. But they have also been observed at the micropylar end. In all cases investigated the aposporous E.S. initials are situated in the cell-layer next to the E.M.C. They can be observed even when the E.M.C. is in the resting stage (v. Table 5). They are characterized by their size, by large nuclei having several deeply stained nucleoli. They soon become separated, as it were, from the nucellar cells. From the records shown in Table 5 it does not appear that the aposporous E.S. initials were observed more frequently while the E.M.C. was at prophase than when it was in the resting stage. In 13 per cent of the cases 2 aposporous E.S. initials were observed in the same nucellus.

The competition between the aposporous E.S. initials and the E.M.C. can be observed at an early stage, already when the latter is at prophase (v. Fig. 19). But it becomes still more evident later. As already pointed out, the real disorganisation most probably takes place during the triad stage or at the beginning of the nucleus division in the legitimate E.S. But we should like to emphasize once more that it is difficult to determine from the material studied what part this competition plays in the disorganisation. In Fig. 23 (= Fig. 29), for instance, the aposporous E.S. initial forces its way up from the chalazal region and seems to push the megaspores out of the way. Only two megaspores are shown in the figure. They are in process of degeneration. The third megaspore is also discernible, but only rests remained. It is therefore possible that all three megaspores were in process of degeneration before they had been influenced by the aposporous E.S. initial. There are indications that at least up to the »tetrad» stage there is a joint action between degeneration and competition. As shown by the above example, degeneration may occur either before or after the reduction division. It might be expected, however, that when a megaspore, as, for instance, that in Fig. 24, has reached a binucleate E.S. stage it would be easily able to compete with the aposporous E.S. initial and produce embryos. As already pointed out, that seldom seems to be the case however. Altogether 46 progeny plants of strain 746 were examined, and here, too, no instances were observed in which the embryo formation had taken place from an E.S. with a reduced chromosome number.

Thus the aposporous E.S. initial is as a rule clearly differentiated when the normal E.M.C. is still in the resting stage. But its first division does not take place until after the tetrad stage, in the cases observed, at the earliest when the normal E.S. has reached the binucleate stage. During this period it continues to grow and before beginning division pushes up, as it were, out of the chalazal region to occupy the place of the normal E.S. (v. Fig. 23). Only final stages of the division of the aposporous E.S. initial were studied, but these stages seem to indicate that the division is somatic. No later stages of the development of the E.S. up to its complete organisation than 4-nucleate stages were obtained.

Strain 703 (Figs. 12—16). — The studied sections of strain 703 were much more difficult to explain than those of strain 746. So far only 17 flowers have been investigated. In these the normal E.M.C. was most frequently at early prophase. Only one case with probable

diakinesis and two with megaspores were obtained. In addition there are several cases in which the E.M.C. could not be found or was in a degenerated form (v. Fig. 12). In the material of 703 studied the E.M.C. was on the whole seldom so well developed and marked as in strain 746. The material conveys the suggestion that the normal E.M.C. degenerates very often, perhaps before or at an early stage of the reduction division. Megaspores however appear to be formed in certain cases, and studies of progenies also indicate that megaspores must occur.

Aposporous E.S. initials were observed in 11 of the 17 flowers, i. e. the same percentage as in strain 746. They were often difficult to find, but the material does not permit a definite answer to the question as to whether the aposporous cells in strain 703 are differentiated later in relation to the development of the E.M.C. than in strain 746. A number of observations, however, seems to indicate that this is so. To sum up, it may be said with respect to the development of the female gametophyte (the terminology for the formation of gametophytes and sporophytes employed here and in the following pages is that introduced by GUSTAFSSON in 1939 a) that: 1) in the sexual strain 709 it arises as a rule from an E.M.C. after the reduction division; 2) that this development is also possible in strains 703 and 746, but that in these strains apospory occurs normally. Thus, as already pointed out, in 703 and 746 the development of gametophytes takes place in the same manner as that described by TINNEY (1940) and KIELLANDER (1941 a) in the apomictic strains.

F. COMPARISON BETWEEN THE DEVELOPMENT OF P.M.C:s AND E.M.C:s IN STRAINS 709, 703 AND 746.

In strain 709 the divisions in the P.M.C. and in the E.M.C. seem to take place about simultaneously in comparisons in one and the same flower. In certain cases they were a little more advanced in the P.M.C. than in the E.M.C. An example or two may be suggestive of this: 1) E.M.C. diakinesis—P.M.C. metaphase—interkinesis; 2) E.M.C. dyad—P.M.C. pollen mature. But such cases as the following have also been obtained: E.M.C. second anaphase—P.M.C. metaphase—interkinesis. In one case, however, a great deviation from the simultaneous development in the E.M.C. and the P.M.C. was observed. The E.M.C. was in prophase, although somewhat difficult to control, while the P.M.C. had developed to the stage shortly after the tetrad formation.

In the few observed cases of strain 703 the development of the

E.M.C. and the P.M.C. appears to follow the same evolutionary rhythm as in strain 709. Strain 746, however, behaves differently in this respect. In this strain the P.M.C. divides much earlier than the E.M.C. When the E.M.C. is in early prophase the P.M.C. has already developed so far that the tetrad formation has been accomplished. That was also the cause why the sections for embryological studies were unsuitable for a study of the division of the P.M.C. Only one exception to this rule was observed, the E.M.C. being in second anaphase and the P.M.C. only in first metaphase. No explanation of this divergency could be obtained.

From the material of strains 709 and 746, however, it is evident that there is a significant difference between the two strains with respect to the developmental rhythm of the E.M.C. and the P.M.C. This difference is of particular interest in view of the fact that strain 746 is apomictic and strain 709 sexual, to which we shall return in another connexion (see p. 107).

G. POLLINATION EXPERIMENTS.

Detailed reports of emasculation and pollination experiments have previously been published (ÅKERBERG, 1936 a, 1939). Here we shall therefore give only a brief summary. Of the 12 strains now being dealt with 10 were included in these experiments. Altogether more than 11,000 flowers were emasculated. About one-fourth of the flowers were not pollinated and therefore, with but two exceptions, produced no seeds. The exceptions were in all probability due to incomplete emasculation (ÅKERBERG, 1936 a). The pollen of *Poa pratensis* and of *P. alpina* have the same effect on the production of seed, whereas flowers pollinated with the pollen of *Poa palustris* and *P. nemoralis* have so far failed to give any seeds. A few solitary seeds have been obtained with *Poa glauca* as the male plant, and with *P. compressa* a relatively small number of flowers were pollinated, but the seed setting was comparatively good. On the whole seed setting in these emasculation and pollination experiments was poor, but that was due to the great technical difficulties involved. From the results obtained in these experiments and others the exceedingly probable conclusion has been drawn that pollination in some way or other is necessary for the development of the ovule in both apomictic and sexual strains of *Poa pratensis*.

Both ARMSTRONG (1937) and TINNEY (1940) mention that they found pollen tubes in the stylar tissue, but neither of them observed

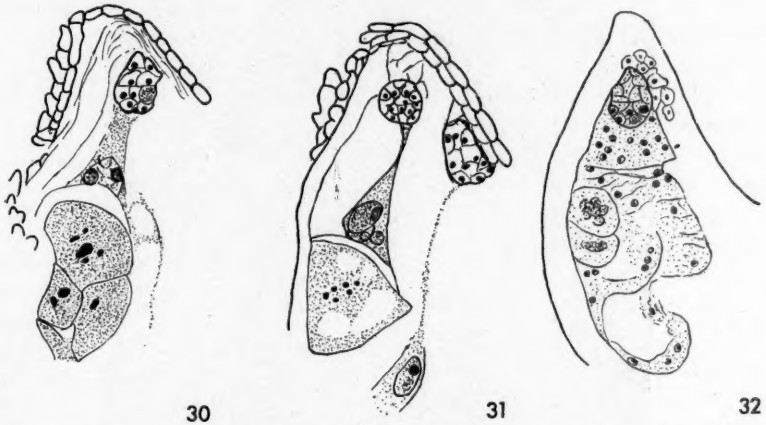
that fertilization had occurred, although ARMSTRONG assumes that to have been the case. This assumption is based on an observation that the embryo was binucleate or multinucleate in fixations made shortly after the stamina had shed their pollen. In one case it was possible to determine the chromosome number in the embryo. Fertilization is assumed to have preceded the divisions of the embryo. In view of TINNEY's results, given below, and ours it is probable that the multicelled embryos observed by ARMSTRONG arose parthenogenetically, for TINNEY points out that the egg develops parthenogenetically into an embryo, and frequently this development begins before the stigma of the ovary is receptive to pollen. Anthesis usually precedes endosperm development, but it has not been shown that the presence of pollen on the stigma is prerequisite to the inception of the endosperm. Hence, while these facts indicate that pollination is unnecessary to embryo-development, its necessity for, or influence on, endosperm-development is unknown. Aborted ovules with no endosperm were observed.

The question as to the real influence of the pollen on embryo and endosperm development in *Poa pratensis* is of great interest, especially in view of the results recently obtained in other pseudogamous species (*Hypericum*; NOACK, 1939; *Potentilla*; GENTCHEFF and GUSTAFSSON, 1940 a). In these species it has been shown that the central nucleus, as a rule at any rate, must be fertilized to ensure the formation of endosperm. TINNEY's investigations show that pollen is not necessary to stimulate the development of the embryo (autonomous parthenogenesis according to GENTCHEFF and GUSTAFSSON). ANDERSEN's material (1927) also seems to suggest this, even if such a conclusion was not drawn. In our material exceedingly small ovules appeared in some cases in emasculated, unpollinated flowers of apomictic types, which may have been weak embryo formations without endosperm. That, too, would indicate autonomous parthenogenesis, at the same time as it would be an indication that the pollen either stimulates or fertilizes the central nucleus.

An investigation of strain 701 seems to prove that that is probably the case. In the summer of 1935 emasculated, unpollinated flowers and emasculated flowers pollinated with pollen of strains 701 and 5303 were fixed. The former, i. e. the unpollinated flowers, were fixed five days after emasculation. The flowers had by then dehisced and the stigmata were quite visible. Eight flowers were investigated. In one of them the embryo-sac was fully organised with an egg-nucleus, but in each of the others there were a many-celled embryo, only one central

nucleus (the polar nuclei had thus fused) and several antipodal cells (see Figs. 30 and 33).

The flowers pollinated with pollen of strains 701 and 5303 have a somewhat more variable appearance. They were fixed 2—4 days after pollination. In all the flowers there were more or less multicelled embryos. Several of them had only one large central nucleus and antipodal cells like the unpollinated flowers. In one case fusion of the polar nuclei was observed, in another (see Fig. 31). a small nucleus



30 31 32

Figs. 30—32. — 30. Embryo, central nucleus and 3—4 antipodes in strain 701. — 31. Two embryos, one of them with two polar nuclei and a male(?) nucleus. — 32. — Embryo, endosperm and degenerating antipodes. — 30. Emasculated, not pollinated flower. — 31 and 32. Pollinated flowers.

close to the polar nuclei, while in three cases endosperm with few or many nuclei was met with (Fig. 32). These four last-mentioned cases do not of course furnish conclusive proof that stimulation or fertilization of the central nucleus precedes and is prerequisite to the development of endosperm and seed in strain 701. But if they are compared with the results of the embryological investigations of unpollinated flowers and of the emasculation and pollination experiments, it seems to be highly probable that induced endosperm formation occurs in this strain, and then probably fertilization of the central nucleus. (On this assumption the occurrence of endosperm in only a relatively small number of the pollinated flowers agrees well with the fact that only a few of the emasculated, pollinated flowers set seeds. It is also possible

that in some cases fertilization did not have time to take place before fixation.) Thus in strain 701 the embryo seems to develop by autonomous parthenogenesis but the endosperm development is induced, i. e. development takes place in the manner described by GENTCHEFF

Fig. 33 = Fig. 30. Microphoto. Strain 701. Embryo, central nucleus and 3—4 antipodes. Emasculated, not pollinated flower. — (Photo O. MATTSSON.)

and GUSTAFSSON (1940 a) in certain species of *Potentilla*. Investigations are at present being carried on to ascertain the extent to which this type of seed formation occurs in other strains of *Poa*.

H. SEED SETTING.

Seed setting in *Poa pratensis* has already been investigated by several workers (FR. NILSSON, 1933, 1934, 1937; ÅKERBERG, 1936 a).

In these reports various cases of modifications and hereditary variation in seed setting after isolation and free flowering have been described and discussed. Several of the strains dealt with in this paper were included in the investigations mentioned. Referring the reader to the data for seed setting in *Poa pratensis* recorded in these works and mentioning that this question will be treated in detail later in a work by NISSEN and ÅKERBERG, we shall give here only a few further examples of modificatory and hereditary variation and shall especially endeavour to show the connexion between seed setting and the development of the female and male reproductive organs.

The material originates from the years 1936—1939. In 1936 and

TABLE 6. *Seed setting after isolation and free flowering.*

	Ä 787	701	702	703	704	5303	746	G 258	768	G 125	709	813
						Percentage of seed after isolation						
1936.....		1,6	9,3	8,5	26,8	23,4	66,6	51,5	29,3	37,3	66,3	18,0
1937.....		1,7	11,1	18,2	18,2	38,8	51,6					
1938.....	26,3	0,9	2,7	4,3	25,7	15,4		55,1	35,5	4,0		
1939.....		1,0					67,4					
						After free flowering						
1936.....		16,3	42,8	53,0	53,4	69,2	81,5		76,2	44,3	78,2	81,3
1937.....		56,4	70,2	71,2	49,1	77,3	86,1					
1938.....	90,5	56,2	54,7	34,2	61,4	67,3		92,9	90,8	95,5		
1939.....		62,2					82,0		75,8		68,0	

1937 two panicles of each plant were enclosed in the same bag, but in 1938 and 1939 the two panicles were bagged separately. In determining the seed setting in 1936 and 1937 200 flowers per panicle were selected, but in 1938 and 1939 all flowers from 25 spikelets were taken. Flowers and spikelets were taken from different parts of the panicle. It has been shown in an earlier work (ÅKERBERG, 1936 a) that both procedures appear to give a satisfactory expression of the seed setting per panicle.

Modificatory variation. — Both FR. NILSSON (l. c.) and ÅKERBERG found that habitat and season may have a considerable influence on seed setting after isolation as well as after free flowering. Moreover, FR. NILSSON (1937) investigated seed setting with a variation of the period of isolation and found that the length of the isolation period has an effect on the yield. A long period of isolation gave few seeds.

Table 6 gives the seed setting of the 12 strains during 1936—1939. The figures recorded originate from one to several plants, all of which were one year old when tested. There is a great variation in the plant

values for one and the same strain, in the first place, with respect to seed setting after isolation, but also after free flowering. To illustrate this the two apomictic strains 702 and 704 have been chosen from the years 1937 and 1938 and 1936 and 1938 respectively, as a rather extensive material of these strains has been investigated. Thus both strains are represented by figures for one year when two panicles of

TABLE 7. *Seed setting after isolation and free flowering in strains 702 and 704 (in 1936 and 1937 the panicles were bagged together, in 1938 they were bagged separately).*

Plant	702—1937				702—1938			
	Isolation		Free flowering		Isolation		Free flowering	
	Panicle 1	Panicle 2	Panicle 1	Panicle 2	Panicle 1	Panicle 2	Panicle 1	Panicle 2
1	16,5	9,5	60,0	55,0	2,0	0,8	25,0	22,7
2	13,5	18,0	72,5	77,5	0,0	0,0	39,3	69,8
3	14,5	11,0	77,0	72,5	7,7	1,1	66,0	74,2
4	15,0	13,0	69,0	60,5	0,0	6,1	61,6	65,7
5	8,5	5,5	74,0	68,5	5,6	2,5	64,5	55,1
6	22,0	15,0	77,5	75,5	4,9	0,8	56,5	55,5
7	2,5	0,0	62,0	69,5				
8	No isolation		64,5	70,5				
9	3,5	6,0	73,0	81,0				
10	13,0	12,5	75,0	66,5				
	704—1936				704—1938			
1	11,5	19,5	68,0	55,0	19,2	34,6	90,9	63,6
2	0,0	0,0	56,5	43,5	29,6	39,2	55,3	56,3
3	46,0	38,5	45,0	40,0	28,6	20,6	50,0	51,4
4	20,0	20,0	57,0	36,5	29,0	28,1	61,8	60,5
5	56,0	42,0	63,5	56,0	13,5	16,9	61,3	62,7
6	36,0	31,5	No free flowering		21,8	27,2	62,6	59,5

each plant were isolated in one bag, and one year when the panicles were isolated separately (Table 7).

The material has been subjected to an analytical examination of the variation (Table 8). If the results of the isolations are studied first it will be found that in strain 702 and in strain 704 the interplant variation is greater than the intraplant variation when the panicles had been bagged together. No such difference was obtained when the panicles had been isolated separately. It is therefore evident that the method of isolation itself may have a great influence on seed setting

in various ways, resulting in a highly variable production of seed by one and the same strain in different isolations. It is a more difficult

TABLE 8. *Analysis of variation of seed setting after isolation and free flowering in strains 702 and 704.*

Year		Degrees of freedom	Square total	Mean square	Variance quotient	P
	Strain 702. Isolation					
1937	Interplant	8	496,00	62,00	7,14	$< 0,01$
	Intraplant	9	78,12	8,68		
1938	Interplant	5	27,61	5,52	0,61	$> 0,2$
	Intraplant	6	54,31	9,05		
	Strain 704. Isolation					
1936	Interplant	5	3344,00	668,80	23,85	$< 0,001$
	Intraplant	6	168,25	28,04		
1938	Interplant	5	396,30	79,26	2,19	$< 0,2$
	Intraplant	6	217,43	36,24		
	Strain 702. Free flowering					
1937	Interplant	9	698,02	77,56	3,83	$< 0,05$
	Intraplant	10	202,62	20,26		
1938	Interplant	5	2593,39	518,68	5,61	$< 0,05$
	Intraplant	6	554,48	92,41		
1937 1938	Inter-year	1	1782,46	1782,46	7,58 [1]	$< 0,05$
	Interplant	14	3291,41	235,10	4,97	$< 0,01$
	Intraplant	16	757,10	47,32		
	Strain 704. Free flowering					
1936	Interplant	4	544,15	136,04	1,62	$> 0,2$
	Intraplant	5	419,75	83,95		
1938	Interplant	5	795,17	159,03	2,51	$> 0,2$
	Intraplant	6	380,74	63,46		
1936 1938	Inter-year	1	464,27	464,27	2,10 [1]	$< 0,2$
	Interplant	9	1339,32	148,81	2,04	$< 0,2$
	Intraplant	11	800,49	72,77		

matter, however, to assign the reason for this variation. But attention should be called to the enormous variation in pollen quality obtained at least in some strain in one and the same year (p. 14).

[1] See text.

It must be regarded as probable that isolation further increases the variation in the quantity and quality of the pollen. There is also a possibility that the culms are easily damaged in isolation, resulting in disturbances in the supply of nutritive material, but these injuries are difficult to detect. The material, however, confirms the result obtained by FR. NILSSON (l. c.) that *Poa pratensis* is very sensitive in its production of seed to the influence of external factors, more sensitive than several other Gramineae. A single isolation therefore does not give a very reliable expression of the seed setting of *Poa pratensis* after isolation.

The analysis of seed setting after free flowering gives somewhat different results for strain 702 and strain 704. In the case of strain 702 there is a very great probability that the interplant variation is greater than the intraplant variation ($1937 + 1938$ $P < 0{,}01$), whereas such a difference does not appear in strain 704. The interplant variation should be an expression of differences in soil, and it may be possible that such differences occurred in the case of the plants of strain 702 but not those of strain 704. Moreover, there is a factor which may possibly explain why no plant differences are discernible in strain 704 in a material of the size treated here. In contrast to strain 702, the seeds of strain 704 loosen very readily and this may have produced a still greater variation in the seed setting of this strain, even if the greatest care was exercised in harvesting the panicles. Among the 12 strains investigated, strain 5303 also showed a tendency to release the seeds easily, although not to the same extent as strain 704.

Table 8, too, shows the results of an examination of the seasonal differences in seed setting after free flowering. For strain 702 the variance quotient has been calculated from $\frac{\text{inter-years}}{\text{interplants}}$, a probability of a seasonal difference between 1937 and 1938 of $0{,}99 > P > 0{,}95$ being obtained. In the case of strain 704 such a difference could not be shown in 1936 and 1938 ($P < 0{,}95$). The variance quotient was calculated, according to BONNIER and TEDIN (1940), from $\frac{\text{inter-years}}{\Sigma \text{ inter- and intraplants}}$. A seasonal difference in the material investigated need not imply a reaction to the weather conditions of the year. The cultivations of the different years were not grown in the same locations and therefore certainly not in the same soil. Further, the plants in the different cultivations may have had varying supplies of pollen in flowering.

Hereditary variation. — The existence of differences in seed setting

after isolation and free flowering, on the one hand, between apomictic and sexual strains, on the other hand, within the apomictic strains and within the sexual strains, is a particularly interesting finding. Unfortunately relatively few data are available for the two sexual strains 709 and 813. But it is evident, however, that both strains are equal to the best of the apomictic strains as regards seed setting in free inflorescences, and in the case of strain 709 also after isolation. For strain 813 a value of 18 per cent is recorded after isolation, but previously between 45 and 65 per cent seed production has been obtained after isolation, hence strain 813, too, seems to be capable of good seed setting with the aid of its own pollen. Thus as far as seed setting is concerned, strains 709 and 813 do not seem to be inferior to the apomictic strains, and do not show any differences among themselves in this property.

Among apomictic strains both FR. NILSSON (1933 and 1937) and I myself (ÅKERBERG, 1936 a) have shown considerable differences in seed setting after isolation and also, though in a lower degree, after free flowering. As pointed out above, isolation in pergamin bags implies in itself a certain reduction in seed production. But the experiments were nevertheless carried out in such a manner that it was possible to show that certain strains set practically no seeds when compelled to rely solely on their own pollen formation, while others, under the same conditions, give a seed setting quite comparable to that after free flowering. Strains 701 and 746 may serve as examples of these two categories. The distribution of the mean values for these two strains, recorded in Table 6, among the plant values is shown in Table 9. The seed setting of the strains was investigated not only after isolation in pergamin bags but also by isolating the plants at wide distances from each other. By the latter method a very reliable measure of the seed production of a plant with its own pollen was obtained (corresponding to self-fertility in sexual strains). In addition to clone plants of strains 701 and 746, maternal progeny plants were also employed, denoted in the table by I_1 and I_2, according to whether they belonged to the first or second progeny generation.

Irrespective of the mode of isolation, no plant of strain 701 yielded more than 10 per cent seeds, distance isolation giving the same seed setting as bagging, I_1 and I_2, as expected, behaving alike. In free flowering strain 701 gives a rather variable seed setting, ranging from 5 per cent to 90 per cent. We shall recur below to this variation in seed setting and to the fact that I_2 gave throughout a high yield of seeds.

TABLE 9. *Seed setting after isolation (in pergamin bags and by distance) and after free flowering in strains 701 and 746.*

Seed setting	Number of plants											
	Isolation				Free flowering			Isolation			Free flowering	
	Pergamin bags			Distance				Pergamin bags		Distance		
	Clone	I_1	I_2	I_1	Clone	I_1	I_2	Clone	I_1	I_1	Clone	I_1
	Strain 701							*Strain 746*				
0,0— 1,0	2	13	3	3								
1,1— 5,0		7	1									
5,1—10,0		1				2						
10,1—30,0					1	6		1				
30,1—50,0					1	6			2			
50,1—70,0						6			4			
70,1—90,0						1	4	1		3	2	6
>90,0												

Strain 746, on the other hand, showed a very good seed setting both after isolation and after free flowering, isolation in pergamin bags, as was to be expected, giving a greater variation than free flowering. It is therefore evident that in strains 701 and 746 we have two undoubtedly different apomictic strains as far as seed setting is concerned.

The cause of this difference is certainly to be found in the first place in the pollen formation. From Table 4 it will be seen that strain 701 has, qualitatively, poorer pollen than strain 746 and probably also quantitatively. The connexion between seed setting in isolation and pollen fertility in *Poa pratensis* has been demonstrated previously (FR. NILSSON, 1933 and 1937; ÅKERBERG, 1936 a). But Table 9 also seems to show that strain 701 has a lower seed setting after free flowering than strain 746. By means of correlation investigations and other tests both NILSSON and ÅKERBERG have demonstrated a difference in seed setting in different strains after free flowering. NILSSON endeavours to explain the poor production of seed in strains like 701 by assuming a defective functional ability in the female sexual cells. In a previous paper (ÅKERBERG, 1936 a) it was shown that a strain giving a low yield of seeds after isolation should also be expected to show a decreased seed setting in free flowering, owing to its being dependent on pollen from the outside. It is of course not inconceivable that a certain difference with respect to the number of embryo-sacs per flower

capable of development may be shown also among apomictic strains. But it is a more difficult matter to assume the existence of a parallelism between that and pollen fertility. It seems to be more probable, however, that the poor seed setting of strain 701 in free flowering is the result of an unsatisfactory supply of pollen rather than an impaired embryo-sac formation. Among the plants of strain 701 investigated in 1938 were the four I_2 and the two I_1 plants recorded in Table 9. The latter were surrounded by strains having a poor pollen quality, while the four I_2 plants were planted in another part of the field, surrounded by at least some strains with a good seed setting after isolation. The I_1 plants gave 6,8 per cent and the I_2 plants 81,1 per cent seeds after free flowering. This difference is probably to be ascribed to a great extent to the supply of pollen. The seed yield of the I_2 plants is so high that we can hardly speak of an impaired embryo-sac development.

Of the other strains included in Table 6, the material of 702, 703, 704 and 5303 was so abundant that these strains can be said to have a poorer seed setting with their own pollen than with pollen from other plants. According to Tables 3 and 4, all these strains were also in one respect or other defective as regards pollen formation. The plants of strain 768 show wide variations, and so do the few plants of strain G 258. Only one plant of strain Ä 787 was investigated.

To sum up it may therefore be said that positive differences between the strains were shown in the production of seeds after isolation by apomictic types of *Poa pratensis*, and that in strains with impaired pollen formation seed setting is more or less reduced, but it may also occasionally (modificatorily) be low in strains with good pollen. The reason for the relatively low seed yield of certain strains after free flowering is for the present less clear. In certain cases, however, it seems to be due to an unfavourable supply of pollen.

I. 1000-SEED WEIGHT.

In an earlier paper (ÅKERBERG, 1938) we showed the great variation in the 1000-seed weight found between different strains of *Poa pratensis*. 126 different strains were examined with a variation ranging from 150—200 mg. to 600—650 mg. The majority of the strains lie between the limits 300 and 500.

The 1000-seed weights of the 12 strains are shown in Table 10 for the years 1936—1938. These values are also based on seeds from one to several plants. A considerable variation is found, ranging from 295 mg. to 630 mg. The five strains Ä 787, 701—704, the chromosome

numbers of which are < 55, have the lowest 1000-seed weight, generally 400 mg. The other strains, having chromosome numbers above 75, have a 1000-seed weight of more than 450, the normal weight of five of the strains apparently being above 500 mg., while four of them have reached values above 600 mg. Thus there is a connexion between chromosome number and 1000-seed weight, but the latter seems also to be influenced to a considerable extent by other factors (cf. 768—38,

TABLE 10. *The 1000-seed weight of the 12 strains during the years 1936—1938.*

	1000-seed weight in mg.											
	Ä 787	701	702	703	704	5303	746	G 258	768	G 125	709	813
1936.....	—	380	375	366	295	504	617	—	460	—	630	604
1937.....	—	396	426	404	386	553	538	—	—	—	—	—
1938.....	365	—	395	330	329	500	—	475	483	625	—	—

2n = 86, 483 mg. and G 125—38, 2n = 87, 625 mg.). It is highly probable that a variation in the number of seeds per panicle or per spikelet also involves a variation in the 1000-seed weight. Five of the six strains for which records of the 1000-seed weight are available for 1936 and 1937 have throughout a higher weight in 1937 than in 1936. Such a seasonal difference in the size of the seeds is of course to be expected and is well known from seed testing stations. The sixth strain, 746, seems to form a noteworthy exception to the others. If we return to the primary figures, however, it will be seen that the weight for 1936 is based on two widely differing values (750 mg. and 484 mg.), one of these values thus being abnormally high for strain 746. This yearly mean value is therefore not quite reliable.

J. GERMINATION TESTS.

The germination tests carried out so far had chiefly in view a comparison between the apomictic and the sexual strains for seeds of different ages. The results of some of these tests are summarized in Table 11. In two experiments (I and III) a comparison was made between 1-year-old and 3-year-old seeds of strain 709 and two apomictic strains, in one experiment (II) only 3-year-old seeds and the apomictic strains were employed, while in the fourth experiment a comparison was made between 1-year-old and 3-year-old seeds of strain 709. The seeds of strain 709 include those obtained after isolation and after free flowering. No difference was found between these two categories of

seeds and therefore they have not been separated in the following discussion. Experiment III, which was performed with the kind assistance of Mr. I. GADD of the Swedish State Seed Testing Station, is reported in detail in Table 12. This experiment also included 4-year-old seeds. The apomictic strains employed were, in addition to strains 701 and 746, also strains 702—704, 5303 and 5305, a strain closely allied to 5303.

According to the tests made so far when the seed is one year old, the germination of strain 709 does not seem to differ significantly from that of the apomictic strains. It is characterized, however, by a con-

TABLE 11. *Germination in 1-year-old and 3-year-old seeds of strain 709 as compared with seeds of apomictic strains.*

Experiment	Strain 709	Germination in percentage after 28 days — Apomictic strains			
		1	2	3	M
			Age of seed: 3 years		1—3
I	52	78	83	—	80,5
II	65	82	77	87	82,0
III	55	79	89	—	84,0
IV	54	—	—	—	
			Age of seed: 1 year		
I	80	73	78	—	75,5
III	63	86	89	—	87,5
IV	92	—	—	—	

siderable variation (63—92 per cent). The state of things is otherwise when the seed has reached the age of 3 years. The apomictic strains still have the same capacity of germination as they had at 1 year of age. (M for the tests was 82,2 and 81,5 per cent respectively as compared with 75 per cent for the State sealing limit for Swedish seeds of *Poa pratensis*.) Strain 709, on the other hand, has a much lower germination (M for the tests being 56,5 per cent). This decrease in the capacity of germination is probably due to a reduced vitality in the zygotes of strain 709. Since the zygote formation in strain 709 takes place from gametes having varying chromosome numbers (cf. p. 18), some of the zygotes should be expected to constitute less vital combinations. Perhaps the endosperm formation, too, is not always quite good.

That the seeds of strain 709 possesses a lower vitality than the apomictic strains is also evident from a study of the rate of germination (see Table 12). In this table a comparison is made between strain

709 and strains 701 and 746. It should be pointed out that strain 746 has approximately the same chromosome number as strain 709, viz. 2n = 80 and 90 respectively. If we first compare the results for 3-year-old and 1-year-old seeds, it will be seen that after 10 days strain 709 had only one or two per cent germinated seeds while strains 701 and 746 had between 26 and 61 per cent. After 20 days there are twice as many seedlings of strains 701 and 746 as of strain 709. The slow rate of germination in strain 709 may of course be partly due to the high chromosome number (corresponding to the connexion between chromosome number and development in a quantitative chromosomal increase almost regularly observed in a species, also in *Poa*, cf.

TABLE 12. *Germination tests in strains 709 compared with strains 701 and 746. Tests carried out at the State Seed Testing Station.*

Strains and age of seeds	Percentage seedlings after number of days					
	10	20	28	38	abnormal	died
709 4 years ...	0	31	45	50	4	46
» 3 » ...	3	41	55	57	7	36
» 1 year ...	1	37	63	74	6	20
701 4 years ...	23	45	53	55	6	39
» 3 » ...	55	75	79	81	4	15
» 1 year ...	58	85	86	89	5	6
746 4 years ...	2	52	69	76	14	10
» 3 » ...	61	87	89	89	4	7
» 1 year ...	26	83	89	90	4	6

MÜNTZING, 1936 and 1940). But a comparison between strains 709 and 746 shows that that is far from being the explanation. A similar slow rate of germination in 709 was also evident in other experiments.

The results for 4-year-old seeds were somewhat different. The experiment conveys the impression that after a storage of 4 years the germination capacity of seeds of even apomictic strains in *Poa pratensis* is greatly reduced. The difference between strain 709 and strains 746 and 701 is not so marked as was to be expected. Further experiments are obviously necessary, and further germination tests would be of interest for other problems, as, for instance, the connexion between chromosome number and rate of germination, the connexion between the germination of the seed and the chromosome number in the endosperm (provided that the endosperm is fertilized). Nothing definite can as yet be said as to the behaviour of *Poa pratensis* in this respect. An answer to this question may possibly be obtained by pollinating

an apomictic »self-sterile» strain, preferably with a low chromosome number, with pollen from strains having widely varying chromosome numbers.

K. POLYEMBRYONY.

In the chapter on embryology it was mentioned that in several investigations of *Poa pratensis* more than one embryo-sac and some cases with more than one embryo had been observed in the same nucellus. In germinating such seeds more than one seedling per seed should be expected, and cases of that kind have also been described. Among the studies on this subject the following are of interest here. [MÜNTZING (1937 a and 1940) especially investigated the occurrence of plants with deviating chromosome numbers among these twin seedlings,

TABLE 13. *The occurrence of polyembryony in strain 709 as compared with strains 702, 703, 704, 746 and 5303.*

Strain	Number of seedlings per seed 1	2	3	4	Total number of seeds	Percentage polyembryony
709	376	8	—	—	384	2,1
702	109	8	1	—	118	7,6
703	91	9	—	—	100	9,0
704	180	39	2	—	221	18,6
5303	213	67	10	3	293	27,3
709	270	8	—	—	278	2,9
746	261	33[1]	—	—	294	11,2

and we shall return to these investigations later.] From two strains KIELLANDER (1941 a) obtained 10 and 15 per cent seeds respectively with twin or more embryos. ARMSTRONG (1937), on the other hand, found no case of twin seedlings in germination experiments on one of the strains investigated. He does not mention the extent of the material investigated, however, and a re-investigation seems to be warranted.

In germination tests with our own material several seeds with more than one seedling were frequently observed. The average for a large number of apomictic strains was 10 per cent seeds with twin or more seedlings (ÅKERBERG, 1939). At the same time it was pointed out that in embryological studies no, or at any rate few, instances of polyembryony were to be expected in a sexual strain, where a tendency to aposporous cell formation is seldom found. In apomictic strains, on the

[1] All seeds with >1 seedling.

other hand, a varying frequency of polyembryony should be expected, due to the occurrence of aposporous cells and embryo-sacs, the capacity of the normal embryo-sac to form embryos, etc. (By way of exception two megaspores may possibly form embryo-sacs; see p. 50). Such a connexion between apospory and polyembryony is especially evident in those cases where the nucellus contains only one spore cell, as in the family Gramineae (SCHNARF, 1929). It has also been shown by ROSENBERG (1930) in the aposporous species of *Hieracium* subg. *Pilosella*.

In the germination tests on which Table 11 is based the occurrence of polyembryony was also investigated. The results are recorded in Table 13, which also gives the values previously obtained for strains 709 and 746. Thus the values given are the total values for seeds from different plants (strain 709, clone plants; other strains, maternal progeny plants). Strain 709 has only 2,1 per cent polyembryony, which is lower than that of any of the investigated apomictic strains ($P < 0{,}01$). Both the values obtained for strain 709 agree very well and, as will be discussed below, it is probable that in polyembryony in this strain one of the seedlings has arisen by apospory. [The frequency of polyembryony in strain 709 is considerably higher than the frequency of twins found in normally sexual plants. MÜNTZING (1937 a) records values between 0,007 and 0,090 per cent for certain varieties of cereals.] There is an evident difference between the apomictic strains in their capacity to form several seedlings. Thus these results seem to be in entire agreement with what was to be expected from the results of the embryological investigations.

Two seedlings per seed is the most common occurrence. In strain 5303, however, quadruplets were observed, and in another material 5 seedlings per seed were obtained in a few cases. In the embryological studies the number of cases with more than one aposporous cell obtained in strain 746 amounted to 13 per cent. The percentage of polyembryony is about 11,2 per cent. An interesting feature is that the figures are of about the same value, even if too great an importance cannot be attached to this correlation. KIELLANDER (1941 a) found, for instance, in a strain of f. compl. *alpigena* an average frequency of uninucleate aposporous embryo-sacs in the same nucellus of approximately three but only 10 per cent seeds with more than one seedling, and ARMSTRONG, as mentioned above, was unable to find that more embryos developed, in spite of frequent cases with two or more embryo-sacs in the same nucellus. It should also be mentioned that in the

investigations of strain 701 mentioned on p. 27 three twin embryos were obtained in 30 controlled embryos.

The two seedlings obtained from one seed frequently appear at different times and in some cases are therefore quite different from each other, even if later they are found to be phenotypically alike. [ANDERSEN (1927) also mentions that in a mature seed both embryos may be fully developed or one of them only partially developed.] It might be expected that in seeds kept for one or two years the frequency of twins would be lower than in seeds newly harvested. In a comparison between 3-year-old and 1-year-old seeds one strain was obtained with a lower frequency of twins for the 3-year-old seeds (3,6 and 14,1 per cent respectively), but this observation was not confirmed by findings in another material containing seeds up to 4 years of age.

L. PROGENY GENERATIONS OF THE SEXUAL STRAINS 709 AND 813.

These progenies merit special attention, firstly for the purpose of studying the inheritance of sexual *Poa pratensis*, secondly for investigating the segregations with other strains, particularly in view of the fact that strains 709 and 813 are morphologically extreme *Poa pratensis* strains and at the same time have high chromosome numbers.

Families of both 709 and 813, obtained by isolation during two generations, I_1 and I_2, have been investigated. Unfortunately only part of the material permits of a comparison between different generations. As a rule these plants were studied in different years. Judging only from the number of survivals after 1 year, the plants planted out did not show a significantly lower degree of viability than plants of apomictic strains, for instance, strains 701 and 746. It must be borne in mind, however, firstly, that the material was not very extensive and, secondly, that a certain selection for vitality might have taken place in planting out. Not all germinated plants were planted out, and those selected were the most vigorous seedlings appearing first (for method of planting out see p. 46).

All these I_1 and I_2 generations were more or less variable in different properties. They varied in height, colour, panicle-formation, vitality, tillering, etc. Some plants did not form panicles, others were extremely weak and gradually died off (see Fig. 34). The variation in colour may be specially mentioned. Both strains are most closely allied to the form complex *irrigata* and are pruinous (a property employed, among others, by LINDMAN in »Skandinaviens Flora» to

distinguish *irrigata* from other form complexes). In the I_1 generation, however, non-pruinous plants occurred.

The morphological variation in the I_1 was confirmed in comparisons between I_2 families. One example of this is shown in Table 14, in which the I_1 and I_2 generations of strain 813 for the same year are compared and also a comparison is made between different I_2 families with respect to plant-height at seed-maturity and the circumference of the tufts, measured in the autumn of the second year (= a measure of the spreading capacity). The I_1 family of strain Ä 756 is both taller and has a superior spreading capacity than the I_2 families of strains

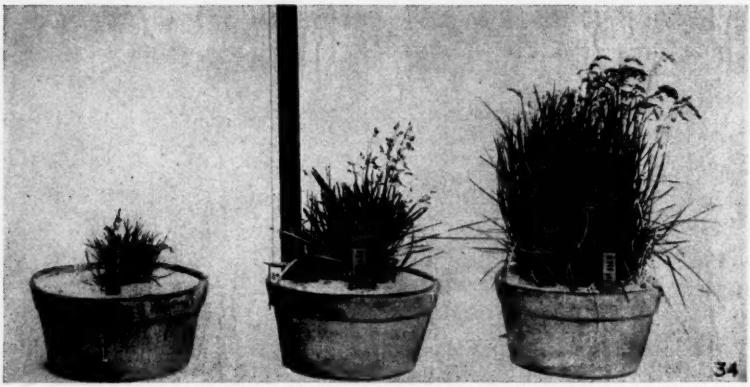


Fig. 34. Strain 813. Three plants from an I_2 family.

Ä 758 and Ä 759. In the material of strains Ä 756 and Ä 757, which consists of only a few plants of each strain, there is no difference between the two generations. No I_2 plant, however, was taller or had a better spreading capacity than the best I_1 plant. A comparison between the I_2 families shows a difference in height between strain Ä 757 on the one hand and between Ä 758 and Ä 759 on the other hand ($P < 0{,}05$), and in spreading capacity between strains Ä 758 and Ä 759 ($P < 0{,}001$), i. e. the three families are differentiated in these properties. We shall return in another connexion to the above material of strains 709 and 813, which is not yet very extensive, and it will then be compared with another similar material (see p. 113).

In three I_1 plants of strain 709 the somatic chromosome numbers were $\pm$ 86, approximately 90 and 88 respectively, i. e. numbers closely corresponding to that of strain 709, 90. Three plants of strain 813 had

$2n = \pm 91$, ± 92 and ± 94, while the somatic chromosome number of a fourth plant was $2n = \pm 130$. As strain 813 was taken from the same population as strain 709 and is morphologically closely in

TABLE 14. *Comparison between the I_1 and I_2 families of strain 813.*

Family No.	Generation	Plant-height cm.	Circumference of tuft in cm.	No. of plants examined
Ä 756.....	I_1	62,2 ± 3,02	107,0 ± 12,21	5
Ä 757.....	I_2	46,7 ± 13,84	100,0 ± 25,15	3
Ä 758[1]	I_2	23,9 ± 3,07	40,9 ± 2,83	10
Ä 759[1]	I_2	26,1 ± 1,88	70,2 ± 4,91	13

agreement with the latter, it seems to be probable, judging from the above chromosome numbers, that its number is about 90—95. The plant with the chromosome number ± 130, which was shorter than the majority of the sister plants but was otherwise not especially weak and

TABLE 15. *Pollen formation and seed setting in I_1 and I_2 of strains 709 and 813.*

Percentage	Number of plants							
	Well developed pollen		Entirely dehisced stamens		Seed setting			
					Isolation		Free flowering	
	I_1	I_2	I_1	I_2	I_1	I_2	I_1	I_2
0— 10				1	10	6	1	2
11— 25		1			3	2	2	1
26— 50	1			1	3		1	3
51— 75		3		3			6	2
76— 90		2					5	1
91—100	6	2	2	2				
Total number of plants examined	7	8	2	7	16	8	15	9

had a good seed yield after free flowering and very good after isolation, probably arose from an unfertilized, likely aposporous egg-cell. Polyembryony occurred in strain 813, as in strain 709, which also renders

[1] One plant of Ä 758 and three plants of Ä 759 did not form panicles, and are not included in the average.

the occurrence of aposporous E.S:s probable (cf. below regarding strain 709).

Table 15 gives a picture of the variation with respect to pollen formation and seed setting. The origin of the material does not permit of a comparison being made between the values for the I_1 and I_2 generations. In spite of the uncertainty incident to every individual determination, it is nevertheless evident that a number of plants were defective as regards pollen formation and seed setting. This is especially true of seed setting after isolation, but several plants gave a low seed yield also after free flowering.

Using strain 709 as the mother plant some *crosses after emasculation* have been made, strains of *Poa pratensis*, *P. alpina* and *P. glauca* being employed as father plants. The F_1 plants obtained are given in Table 16. In all cases, except one, it is evident from the chromosome number of the F_1 plant that a reduced female gamete has functioned. The exception, $2n = \pm 108$, certainly arose from an unreduced female gamete, for according to other crosses the chromosome number of the *Poa alpina* plant employed was in all probability about 40. The occurrence of polyembryony in strain 709 mentioned above led to the assumption of a certain degree of apospory also in this strain. In that event it should also be possible for embryos to arise from unreduced egg-cells, provided the aposporous E.S. initials divide mitotically, as in the apomictic strains of *Poa pratensis*. As such unreduced egg-cells have now been shown, that may confirm the hypothesis put forward. It cannot of course be excluded that the embryo-sac from a legitimate E.M.C. may contain the somatic chromosome number by divisions as in the case of diplospory. But there is nothing in our investigations or those of other authors to indicate that such a division takes place in *Poa pratensis* (cf. also p. 49). Thus the crosses and isolations performed may for the present be said to agree with the embryological finding that in strain 709 the female gamete in the embryo-formation mostly consists of an egg-cell with a reduced chromosome number. This occurrence is not contradicted by the results of the studies of polyembryony. The hybrids with *Poa alpina* and *glauca* will be discussed later (see p. 91). The haploid plant obtained after pollination with the pollen of *Poa glauca* was very weak and died before it could be examined.

The only other F_1 plants studied somewhat closely were the three plants having strains 702 and Ä 694 as father plants, and of these the only F_2 material was from the crossing with strain 702. In both crosses

strains belonging to widely different form complexes from that of strain 709 were used. Strain 702 is closely allied to the *angustifolia* complex, while strain Ä 694 resembles the *alpigena* complex. The F_1 plant of the cross 709 × Ä 694 with $2n = \pm 76$ was with respect to its morphological characters intermediate between the parent plants. Its seed setting after isolation and after free flowering was good. The two plants of the cross 709 × 702, the chromosome numbers of which agreed rather well (plant 1 having approximately 72 and plant 2 between 65 and 70), differed greatly morphologically. In several pro-

TABLE 16. *Chromosome numbers in F_1 and F_2 of crosses with strain 709 as mother plant.*

♂ Strain	2n	♀ 709 2n = ± 90 F_1 plants 2n = ± 1	2	3	4
702	50	ca. 72	65—70		
Ä 694	68	76			
Ä 217	?	64			
Poa alpina I	?	70	66	70	60
Poa alpina II	?	108			
Poa glauca	?	42 (haploid)	ca. 71		

Cross	F_1 2n = ±	F_2 plants 2n = ± 1	2	3
709 × 702	ca. 72	74	72	
» × »	65—70	68	64	67

perties, panicle-formation, earliness, plant 1 resembles strain 709, while plant 2 was very similar to strain 702. Both plants gave a good seed yield after free flowering, in isolation plant 1 yielded 14,3 per cent and plant 2 42,0 per cent.

The morphological difference found in the F_1 was verified in the F_2. The chromosome numbers of some of the F_2 plants were counted. In the F_2 of plant 1 the numbers 72 and 74 were obtained and in the F_2 of plant 2 the numbers 64, 67 and 68, values indicating some difference between the two families. Of morphological characters, the height was measured in both families, the F_2 of plant 1 giving $35{,}1 \pm 2{,}26$ and the F_2 of plant 2 showing $47{,}1 \pm 1{,}51$ with a diff. $= 12{,}0 \pm 2{,}68$ (the probability of a diff. $P > 0{,}999$). Investigated F_2 plants of plant 1 showed a somewhat lower seed setting than F_2 of plant 2. The F_2 plants of plant 1 showed variability in, among other properties, leaf-colour and leaf-breadth, while the F_2 plants of plant 2 were strikingly constant,

all plants being similar in type to strain 702. To obtain an answer to the question as to whether this constancy depends on a certain degree of apomictic seed formation in plant 2 further investigations will be necessary. It can already now be said that in germination experiments 10 per cent polyembryony was obtained in plant 2 and nil in plant 1. These two families do not furnish much information of the genetic difference between strains 709 and 702. But it appears as if each strain possesses a complex of factors dominating in their respective F_1 plants. This variable F_1 may be the result of the heterozygoty of the mother plants and different numbers and combinations of the chromosomes of the female and the male gametes.

Summing up the results of the investigations carried out so far in the sexual strains 709 and 813, the following points may be mentioned: Both strains have sexual propagation but with a tendency to apospory. The progeny arising by self-fertilization is, if subjected to a natural selection, probably intensely selected, for a great variation in the vitality and fertility of the zygotes was observed from the embryonic stage right up to the fully developed plant. Intense inter-crossing must be expected to occur if the progeny grows in the vicinity of other *pratensis* strains. Inter-crossing with strains of the form complexes *angustifolia* and *alpigena* seems to be quite possible.

M. PROGENY GENERATIONS OF THE APOMICTIC STRAINS Ä 787, 701—704, 5303, 746, G 258, 768 AND G 125.

Method. — The seed was germinated on a bell jar apparatus in a seed testing station. A number of the seedlings in a suitable stage of development were placed in small pots, one seedling in each pot. The potted seedlings were those first appearing and it is therefore possible that in some cases a certain amount of selection was made, assuming that the aberrants mentioned below show a different course of germination from that of the normal plants. The material does not permit a thorough analysis of the effect of this selection with respect to the frequency of aberrants obtained. Some experiences, however, go to show that this effect was not great except perhaps in an isolated case, an instance of which will be given below. In support of the view that selection has no great influence on the frequency of aberrants we may mention the material of strains 701 and 702. A rather extensive material of these two strains is available, obtained after emasculation and pollination. *All* seedlings obtained of these strains were planted out. In a comparison made between the aberrants occurring and the

frequency of aberrants in other material planted out no evident difference was found. On that account and in view of the fact that all strains when planted out were treated in exactly the same manner, the comparisons in aberrant frequency made below seem to be justified.

With but few exceptions, the material was fixed while the plants were still growing in the pots. From 1937 the twin plants were also divided and transplanted before fixation. The plants were then planted out in suitable weather in the summer. The plants could be studied to a certain extent in the year they were planted out, but the chief investigation could not be made until the following year, as plants of *Poa pratensis* do not form panicles until the first year after being planted out. The 1935 and the 1936 materials and also to a certain degree the 1937 material could be studied for at least another year.

The morphological analysis of the plants presupposes a good and uniform development of the material. The material planted out at Weibullsholm in 1936 and 1937 in particular satisfied the exacting demands made on it in this respect, as shown by the good agreement between the morphological analysis and the chromosome determination (see below). The most satisfactory results of the morphological investigations were obtained up to the flowering stage, but were controlled by a study of the sterile shoots in the autumn after harvesting the seeds.

The morphology and chromosome numbers of the progeny plants. — As mentioned in the introduction, previous investigations have already shown that the 10 strains Ä 787, 701—704, 5303, 746, G 258, 768 and G 125 have mainly apomictic seed formation. All the progeny material of these strains tested is summarized in Table 17. Altogether 631 plants obtained after isolation, free flowering and crossing with *pratensis* and *alpina* have been studied. Of these 631 plants 562, or 89 per cent, were morphologically similar to their respective mother plants, and in 44 of them an agreement in the chromosome number was ascertained. Some of the 562 plants, especially a number of doubtful cases, have been controlled by investigation of further progenies. Of the remaining 69 plants 50 were haploids or hybrids of *pratensis* and 10 hybrids with *alpina*. The remaining 9 plants (amounting to not more than 1,4 per cent of the entire material) consist of morphologically uncertain aberrants, which have not yet been controlled by progeny or chromosome counts. All the 50 *pratensis* aberrants differed widely morphologically from the mother strains. Chromosomal variability was always accompanied by morphological variability, but as will

TABLE 17. *Progeny investigations of the apomictic strains Ä 787, 701—704, 5303, 746, G 258, 768 and G 125.*

Strain	Maternal plants		Haploids and hybrids in *pratensis*						Hybrids with *alpina*			
	Number	Chromosome number examined	Chromosome number examined			Sure morph. divergence 2n unknown	Total 4—7	Uncertain morph. divergence 2n unknown	Chromosome number examined		2 n unknown	Total 10—12
			hapl.	dipl.	tripl.				dipl.	tripl.		
1	2	3	4	5	6	7	8	9	10	11	12	13
Ä 787.....	3	1	—	—	—	—	—	—	—	—	—	—
701.....	113	7	—	1	1	6	8	—	1	2	2	5
702.....	145	15	—	7	5	1	13	2	1	—	1	2
703.....	46	6	—	3	1	—	4	—	—	—	—	—
704.....	99	5	2	3	11	4	20	—	—	—	—	—
5303.....	66	2	—	1	1	1	3	1	1	—	—	1
746.....	44	1	—	—	—	—	—	—	—	2	—	2
G 258.....	6	2	—	—	—	—	—	—	—	—	—	—
768.....	35	4	—	2	—	—	2	4	—	—	—	—
G 125.....	5	1	—	—	—	—	—	2[1]	—	—	—	—
Total.....	562	44	2	17	19	12	50	9	3	4	3	10

[1] 2n known but does not differ for certain from G 125.

be shown below the latter need not always be accompanied by a change in the chromosome number. The 10 *alpina* hybrids, which will be treated in a later chapter, differed widely from the mother plants. As the measurements, etc. of the maternal progeny plants are included in the previous description of the strain, they will not be dealt with here.

In an earlier paper (ÅKERBERG, 1939) it was shown that the progeny of the facultatively apomictic strains of *Poa pratensis* seems to arise in four different ways, judging from the chromosome number. Such progeny types have been obtained by MÜNTZING (1937 a, 1938 and 1940) and partly by SKOVSTED (1939) and KIELLANDER (1941 b). The first type consists of the maternal plants, having the same chromosome number as the mother plant. The embryological investigation seems to have demonstrated that the apomixis previously shown in these plants consists of apospory attended by parthenogenesis.

The other three types are found among those differing morphologically from the mother plants. In one of these types, designated triploid aberrant in Table 17 and later, the chromosome number is about 50 per cent higher than that of the mother plant. Such triploid types were observed very frequently. About 50 per cent of the chromosomally controlled hybrids recorded in the table are triploids. MÜNTZING (1938) investigated twin aberrants and found about 70 per cent triploids. (This figure is rather high, being based exclusively on chromosomal aberrants, a category that does not include all aberrants.) Among 31 twins SKOVSTED (1939) found 14 pairs, one of the members of which was triploid. It has been assumed that these triploids arose by the fertilization of an unreduced egg-nucleus. The possibility of an unreduced pollen grain having fertilized is contradicted by triploids from crosses between strains with known chromosome numbers [e. g. $♀ 2n = 50 \times ♂ 2n = 90$ gives F_1 $2n = 95$ $(50 + \frac{90}{2})$ and not 115 $(\frac{50}{2} + 90)$]. Besides, no giant pollen grains were met with in pollen investigations.

Embryological studies have shown that an E.S. with an unreduced egg-nucleus normally arises in the apomictic strains from the aposporous E.S. initials, and the most plausible assumption is that these are fertilized. As pointed out above, the embryological investigations did not reveal such a division of the legitimate E.M.C. as to enable an E.S. with the somatic chromosome number to arise. The fertilization of the aposporous E.S:s is also supported by certain investigations of

twins. MÜNTZING (1937 a) reported a twin pair, both members of which had different chromosome numbers from that of the mother plant (mother plant $2n = \pm 52$, twins $2n = \pm 65$ and ± 80). There does not seem to be any possibility of explaining this case except by assuming the fertilization partly of an embryo-sac with a reduced egg-nucleus (zygote $2n = \pm 65$), partly of an embryo-sac with an unreduced egg-nucleus (zygote $2n = \pm 80$), the latter originating from an aposporous E.S. initial. ÅKERBERG (1939) and SKOVSTED (1939) give further examples of the fertilization of two embryo-sacs from the same nucellus, one of the embryos, at least, being triploid.

In the cases mentioned above it is not probable that the two embryo-sacs arose from two megaspores. In that event one of the megaspores would have twice the chromosome number of the other. Generally speaking, it has not been possible to show morphologically any tendency in more than one megaspore to form an embryo-sac. The non-functioning megaspores soon degenerate, and this applies in the first place to the micropylar dyad cell that fails to divide. There is, however, a case obtained in an investigation of twins, not yet fully proved, which may possibly indicate that two megaspores formed an embryo-sac each. An alternative interpretation, however, is the formation of two embryos from two nuclei in an E.S. with reduced chromosome number. A twin of strain 704 ($2n = 53$) was obtained, both members of which had $2n = \pm 29$ and $2n = \pm 58$ respectively. The former differed widely morphologically and evidently arose by *haplo-parthenogenesis*. The latter was recorded several times as being morphologically different, even if with some hesitation occasionally. Further, control determinations in several roots show that in all probability it has a different chromosome number from that of the mother plant, but it is not a triploid. It should thus have arisen by the fertilization of a *reduced* egg-nucleus. This interesting case will now be controlled by testing the next generation.

There is not much to be said regarding the mode of origin of the diploid and haploid aberrants. As a reduced chromosome number is shown only by the legitimate embryo-sac, the former certainly arise by fertilization and the latter by haplo-parthenogenesis. It might of course occasionally happen that the former obtain the same chromosome number as the mother plant, but differ morphologically owing to a difference in chromosome complement. The following scheme (Scheme 1) shows the formation of gametophytes and sporophytes in

apomictic strains assumed above. Strain 704, from which all four types have been obtained, is taken as an example.

Scheme 1. Strain 704, 2n = 53.

	Vegetative cell		Archesporial cell	
Gametophyte-formation	Apospory	Apospory	Reduction division	Reduction division
Sporophyte-formation	Diplo-parthenogenesis	Fertilization	Fertilization	Haplo-parthenogenesis
Number of plants	49[1]	9	3	2
Average 2n number	53	80	54	27
Names in Table 17 and text	Maternal plants	Triploid aberrants	Diploid aberrants	Haploid aberrants

In the embryological investigations autonomous parthenogenesis was shown in the apomictic strains, i. e. a multi-celled embryo may be formed without pollination and has been formed on pollination even before the pollen tube has grown down through the style. This takes place normally in the case of the egg-nucleus in the aposporous embryo-sacs. But nothing conclusive can be said as yet as to the behaviour of the egg-nucleus in the legitimate E.S. According to Table 17 it seems to be fertilized more frequently than to develop into a haploid and should therefore, quite naturally, have a later development than the aposporous E.S. But this result may also be due to the phenomena of selection.

There remains, however, the question as to how an aposporous egg-nucleus ever manages to get fertilized in time. TINNEY (1940) mentions that occasionally the egg-nucleus does not begin to divide until after pollination and that in such cases it has time to become fertilized. As mentioned above, in the twins one of the members is considerably later in development, and consequently should have time to be fertilized. On that account TINNEY (1940) assumes that triploids should be found more frequently in the weaker than in the more vigorous twin. No material has been submitted furnishing positive confirmation of this assumption, but some experiences seem to indicate that it is correct. In 1936 only 20 plants of the 73 seedlings (= 27 per cent of the total) of strain 704 obtained from seeds after isolation were planted out, the first seedlings appearing being selected. All these plants were maternal. In 1937, of 117 seedlings, also from seeds after isolation, 73 plants (= 62 per cent of the total number of seedlings)

[1] 2n was determined in only one of these plants.

were planted out. Among these no less than *14 aberrants* (19 per cent) were obtained, 10 of which were triploids. Ten twin pairs were planted out, among which there were 7 aberrants (35 per cent). We find here a marked tendency to a higher frequency of aberrants in 1937 than in 1936, i. e. in the year the greater percentage of plants were planted out, consequently of later appearing seedlings (probably arisen from late developed embryo-sacs). This is a more plausible explanation than an attempt to interpret the difference as a pure seasonal difference. There is also a tendency to a higher frequency of aberrants in the twins than in the single plants in 1937 (P in both cases for uncorrected χ^2 was $< 0{,}05$). Another striking feature is that strain 704 in addition to a high frequency of twins (Table 13) also shows a high frequency of triploids among the aberrant plants (Table 17). A tendency to a higher frequency of aberrants in twins has also been reported previously (ÅKERBERG, 1939). The difference, however, was not significant.

To enable fertilization to take place it is therefore necessary for the pollen tube to have sufficient time to grow down the style before the egg-nucleus has commenced dividing. According as the male gametes show different rates of growth, for instance, owing to different chromosome numbers, a more intensive selection should be expected among them. It is then conceivable that the aberrant frequency would be influenced by the father plant, provided the pollen grains from different father plants had different rates of germination and growth. Certain crosses have been said to indicate this (ÅKERBERG, 1939). In a further investigation of that material, a cross between strain 701 and *Poa alpina*, it was found that a zygote selection was the more probable explanation. The hybrid *Poa pratensis* × *alpina* germinates much more rapidly than pure *pratensis* (see p. 100). If only plants from the first seedlings are selected, a higher frequency of hybrids should be expected among them than if all seedlings were planted out.

Thus there is at present no sure case in which the *father plant* has been shown to influence the frequency of aberrants. An aberrant frequency varying with the *mother plant* has, however, been obtained in certain material (ÅKERBERG, 1939). Table 17, too, gives an impression of a varying frequency of aberrants. The figures cannot be directly compared, however, on account of the origin of the material. If absolutely comparable material is examined, no difference in frequency can be shown in these strains, which does not, however, contradict the results of the 1939 material, which included other strains.

The total *pratensis* material investigated conveys *the definite impression of a variable aberrant frequency.* In one strain, of which a rather extensive material was available, the frequency of aberrants seems to be practically nil. This problem, however, seems to merit a more comprehensive study than that afforded by this material. For that purpose it is necessary to have a material raised under different condition from that treated here. We shall then also investigate the relation between haploid, diploid and triploid aberrant plants, the connexion between the frequency of twin plants and aberrant types, etc. The total aberrant frequency alone does not furnish a reliable measure of the degree of apospory. Regard must also be paid to the occurrence of triploid aberrants. One of the difficulties of these investigations is that an extensive material is necessary in order to obtain reliable frequency figures.

N. THE MORPHOLOGY, ETC. OF HAPLOID, DIPLOID AND TRIPLOID ABERRANTS.

Haploid aberrants. — Only two haploid aberrant plants were obtained and both of them from strain 704 after isolation. The chromosome numbers were $\pm$ 24 and $\pm$ 29 respectively. Both plants morphologically differed widely from normal *Poa pratensis*, the one having $2n = 29$ showing the greater difference. The latter had a single panicle with spikelets closely compressed against the tips of the panicle branches. The other plant developed a few panicles, the spikelets of which were also somewhat compressed and deeply anthocyanin coloured. Both plants died in the second year. No seeds were obtained from either plant after isolation or free flowering. Progenies of haploid aberrants, however, are known and have been studied (MÜNTZING, 1940; KIELLANDER, 1941 b). The haploid plant described by MÜNTZING had $2n = 36$ (mother plant $2n = \pm 72$). Meiosis was almost completely regular, but the pollen fertility and the seed setting were poor. The progeny was very variable, hence the haploid plant was evidently sexual. The other haploid plant found by KIELLANDER had $2n = 18$ (mother plant $2n = \pm 72$) and was variable in the progeny too.

Diploid aberrants. — The 17 diploid aberrant plants obtained should be divided into two categories, those arising after isolation and those arising after crossing. Ten plants of the former category were obtained and 7 of the latter.

The 10 aberrant plants obtained after isolation originate from 5 different strains and have been compared in several characters with

the respective mother strains. The chromosome numbers are given in Table 18. These numbers are characterized by a certain variation round the number of the mother plant, and this was to be expected inasmuch as the gametes obtain different chromosome sets owing to the irregularity of the reduction division. In this respect the aberrants behave in the same manner as the offspring of sexual species with varying chromosome numbers in the gametes.

Several morphological characters have been compared. Generally speaking, it can be said that the diploid aberrants appear to be vegetatively weaker on the average than the mother plants but nevertheless

TABLE 18. *Chromosome numbers of diploid and triploid aberrants obtained after isolation.*

Strain			Aberrants 2n = ±									
No.	2n =	Type	1	2	3	4	5	6	7	8	9	M
702	50	diploid	48	47	51	51						49
		triploid	75	73	71	72—75						73
703	50	diploid	54									54
		triploid	—									—
704	53	diploid	50	54	58							54
		triploid	68—70	72	80	80	80	81	83	85	87	80
768	80	diploid	85									85
		triploid	—									—
5303	78	diploid	75—80[1]									75—80
		triploid	110—115[1]									110—115

show a marked variation in decrease of vitality among themselves. They are decidedly shorter, have shorter panicles and frequently inferior tillering. In three aberrants the height was measured in two successive years. The height was throughout less in the second year than in the first. On the other hand, no significant difference could be found in leaf-breadth and leaf-length. There is nevertheless a tendency to a decrease also in these properties (see Table 19). Seed setting after isolation and after free flowering is on an average lower in the aberrants than in the mother plants. That cannot, however, be regarded as being due to vegetative impairment alone. As will be shown later, these aberrants seem as a rule at least to be sexual, which may also influence the seed setting. In the few cases in which pollen fertility was investigated, superior as well as inferior pollen quality to that of the mother plants was found. So far very poor pollen has been

[1] Obtained from the chromosome numbers in F_2 plants.

obtained in only one case. In this case and in another in which the pollen has not yet been examined the seed setting in isolation and in free flowering was practically nil. The 1000-seed weight was on the average lower in the aberrant plants, in one or two cases very much lower.

It appears as if these aberrants behave in many respects like the offspring of the sexual strains 709 and 813 (cf. p. 41 et sqq.), which was of course to be expected. The decrease in vitality in all these aberrants is probably due to the complicated inheritance that the cytological investigations seem to indicate should take place. An

TABLE 19. *Comparison between diploid aberrants and mother plants.* (*Aberrants obtained after isolation.*)

	Mother plants. Average value	Dipl. aberrants Average deviation	n	P is recorded if $P \leqq 0{,}05$
Height	74,6 cm.	$-17{,}57 \pm 3{,}89$	9	$P < 0{,}01$
Panicle-length	11,5 »	$-2{,}20 \pm 0{,}435$	9	$P < 0{,}001$
Apical culm leaf: Length	6,4 »	$-0{,}67 \pm 0{,}439$	9	
Breadth	0,40 »	$-0{,}036 \pm 0{,}026$	9	
Penultimate leaf: Length	9,4 »	$-0{,}59 \pm 0{,}621$	9	
Breadth	0,49 »	$-0{,}022 \pm 0{,}026$	9	
Leaf of sterile shoots: Length	23,9 »	$-5{,}14 \pm 2{,}62$	9	
Breadth	0,36 »	$+0{,}006 \pm 0{,}022$	9	
Seed setting: Isolation	17,4 %	$-12{,}36 \pm 3{,}79$	9	$P < 0{,}02$
Free flowering	60,9 %	$-39{,}27 \pm 5{,}93$	10	$P < 0{,}01$

apomictic strain found in nature with, for instance, $2n = 50$ is quite certainly a selected strain and has in some respects a balanced chromosome set. Judging from the cytological results, in a sexual progeny of such a strain many disturbances should occur with an accompanying decrease in vitality, caused by the absolute chromosome number, the chromosome set, the degree of aneuploidy, genic effects, etc. Certain parallels may be drawn here to the progeny of sexual *Poa alpina* (MÜNTZING, 1940).

In 4 of the 7 diploid aberrants obtained from crossing the chromosome numbers are known for certain from the mother and father plants and the aberrant. Only these 4 aberrants will be discussed here. Scheme 2 shows the origin and seed setting of the aberrants.

Using strains 702 and 703 as mother plants and strain 5303 as father plant three aberrants were obtained, the chromosome numbers

of which differ widely from each other (cf. $\frac{50+78}{2}=64$). This again shows the great chromosomal variation existing in the gametes. The three aberrants also show great differences in seed setting, which was determined in two different years. The differences cannot therefore be attributed to modificatory variation. Strain 5303 had a somewhat higher seed yield than strains 702 and 703 (see Table 6), and the variation in seed setting shown by the aberrants is in all probability to be ascribed to the different chromosome sets they received from the parents. The seed setting in one aberrant indicates that in this property it was superior to both parents. The two aberrants from 702 × 5303 differed widely morphologically from one another. The F_1 of the cross

Scheme 2. The origin and seed setting of the aberrants.

♀		♂		Aberrant		
Strain	2n	Strain	2n ±	2n ±	Percentage seed setting	
					Isolation	Free flowering
702	50	5303	78	70	52,8	82,3
702	50	5303	78	66	18,8	60,5
703	50	5303	78	59	0,0	68,5
768	86	708	64	72	0,0	47,3

768 × 708 was interesting inasmuch as it afforded the possibility of studying the inheritance of a qualitative characteristic. Strain 768 is practically devoid of anthocyanin in the panicle, while strain 708 is deeply coloured with anthocyanin. The F_1 was almost as deeply pigmented with anthocyanin as strain 708, and in the F_2 a marked segregation in this characteristic was obtained.

Triploid aberrants. — These aberrant plants will also be divided into two categories, 1) arising after isolation and 2) after free flowering. No less than 9 of the 15 aberrants belonging to the former category were derived from strain 704, and 4 aberrants were obtained after free flowering.

Investigations of the morphological characters of triploid aberrants as compared with the mother plants have already been performed (NISSEN, 1937; MÜNTZING, 1940). The triploid plants in both these studies were not obtained after isolation, but the chromosome numbers nevertheless seem to point to their having been autotriploids. NISSEN

investigated the length of the stomata cells and found that it was greater in the triploid aberrants than in the mother plants. MÜNTZING studied 7 aberrants derived from twin plants. Six of the mother plants had a 2n between 48 and 52, while the seventh had 2n = 68. As several of the characters investigated by MÜNTZING are comparable with those

TABLE 20. *Comparison between triploid aberrants and mother plants. (Material: Aberrants obtained after isolation from 704 and aberrants described by* MÜNTZING, *1940).*

	Aberrants from 704			MÜNTZING's aberrants (n = 7)	
	Average deviation	n	P (recorded when P ≦ 0,05)	Average deviation	P (recorded when P ≦ 0,05)
Height	−11,0 ±3,73 cm.	8	< P 0,05	− 2,56 ±3,41 cm.	—
Panicle-length	− 1,32 ±0,748 »	9	—	+ 1,03 ±0,568 »	—
Apical culm leaf:					
Length	− 0,111 ±0,360 »	9	—	+ 0,004 ±0,118 »[1]	—
Breadth	+ 0,056 ±0,023 »	9	P < 0,05	+ 0,045 ±0,011 »[1]	P < 0,01
Penultimate leaf:					
Length	− 1,11 ±0,356 »	9	P < 0,02	—	—
Breadth	+ 0,016 ±0,023 »	9	—	—	—
Straw-thickness	+ 0,031 ±0,077 mm.	9		+ 0,150 ±0,046 mm.	P < 0,02
Leaf-thickness	—	—		+ 0,386 ±0,071 »	P < 0,01
Vegetative leaf:					
Length	− 4,44 ±0,983 cm.	9	< P 0,01	—	—
Breadth	+ 0,044 ±0,029 »	9	—	—	—
1000-seed weight	+71,5 ±17,33 mg.	4	P < 0,05	+ 75,86 ±31,25 mg.	P very nearly 0,05

measured in our own material, the results are presented together in Table 20.

It is possible, however, that the lack of complete agreement between MÜNTZING's results and our own is due to certain factors. Our material originates from a single strain, 704, and the measurements were made during one single year. MÜNTZING's results are based on several strains, and the plants were measured in two successive years. MÜNTZING mentions that the triploid twin aberrants develop more slowly than the sister plants. It does not seem to be probable that this

[1] See text.

is the result of the high chromosome number alone (cf. p. 77). From our own material it was quite evident that the tufts of the triploid aberrants did not grow as quickly as the maternal plants. After one year the difference in circumference amounted to $-37{,}57 \pm 7{,}23$ cm. This inferior spreading capacity of the triploid aberrants cannot be exactly compared with the low spreading capacity characteristic of the *angustifolia* types. Besides, the latter types frequently seem to be low chromosomal. It is possible that this difference in development may have had a certain influence on at least some characters, so that a comparison between material of different ages is not quite correct. Moreover, the environments of the plants were not the same in both cases. In his measurements of the leaf-breadth and leaf-length MÜNTZING has not distinguished the different straw-leaves. The values are given in the table in the line for the apical straw-leaves.

Judging from our own material, the different leaves of the culm and the vegetative leaves do not seem to react quite in the same manner to the chromosomal increase. The apical leaf, which is the shortest, shows a rather evident increase in width in this material, but no such increase could be shown in the penultimate leaf and the vegetative leaf, even if there is a tendency to increase. On the other hand, these two leaves show a marked decrease in length, which is not apparent in the apical leaf. The increase in leaf-breadth is very striking in MÜNTZING's material, whereas no decrease in length is discernible. As regards the 1000-seed weight there is a good agreement between the *average* differences. But in MÜNTZING's material the difference between triploid and diploid showed a much greater variation than in our material. With regard to height and straw-thickness there is an evident difference in one case, but not in the other, whereas the panicle-length does not show any evident difference in either case.

Even if both investigations showed that an increase in the chromosome number evidently effected certain morphological properties, it is nevertheless difficult to give a general idea of this influence. MÜNTZING points out (1940, p. 160) that »morphologically it is rather difficult to distinguish 'diploid' and 'triploid' *pratensis* twins from each other, and even if the two twins are dissimilar in appearance, it is sometimes difficult to decide which of the two plants has the higher chromosome number». It is therefore probable that more extensive measurements of these properties are necessary in order to eliminate the »intra-variation» and to obtain definite information of the »interplant» differences. It should also be borne in mind that there is hardly any reason

to expect the same evident and uniform effect of an increase in the chromosome number in *Poa pratensis* as in sexual species. This is partly associated with the high chromosome number itself, partly with the fact that the triploid aberrants are not autotriploids in the same manner as those derived from a sexual species. The chromosomal increase in the former is not only quantitative but also to a great extent qualitative, owing to the different chromosome sets in the gametes.

Fig. 35. Strain 704. To the right a normal plant, to the left a triploid aberrant.

However, if we compare the material under discussion with measurements of collected strains having varying chromosome numbers, we shall be able to obtain several fair parallels. Such parallels will be of great interest when we come to discuss the selective value of triploid aberrants (see further p. 116).

The triploid twins in MÜNTZING's material had throughout better pollen quality than the sister plants. Such an improvement was also obtained in our own material, for instance, in four triploid aberrant plants from strain 702. The aberrant plants from strain 704 behaved somewhat differently. In two of the latter no pollen was obtained and

the stamens were small and shrivelled (probably diplontic sterility), in three the pollen quality was distinctly inferior to that of strain 704 (possibly the combined action of haplontic and diplontic sterility). Like strain 704, three aberrants had fairly normal pollen. Seed setting in isolation runs parallelly with the pollen quality, varying from nil to the same as that of the maternal plants. Seed setting after free flowering is on an average somewhat lower in the aberrants, but here, too, the fact that the aberrants seem to be sexual has an effect on the result. In some cases, however, diplontic sterility was probably the cause of the inferior seed yield.

The triploid aberrants from strain 704 show a rather great variation in chromosome number (Table 18). There does not seem to be any correlation between this variation and the properties discussed above. On the assumption that 2n in strain 704 is 53, in the extreme cases $\pm$ 72 and $\pm$ 87 gametes with $\pm$ 19 and $\pm$ 34 chromosomes should have functioned. We have here a similar variation to that in the diploid aberrants. The pollen grain having $\pm$ 19 differs by 8 from the »haploid» number of $\pm$ 27 found in the mother plant. The haploid plant found by KIELLANDER (see p. 53) had 2n = 18, i. e. half the »haploid» number, $\pm$ 36, of the mother plant. It represents a still more marked extreme value, but these cases are possibly only different degrees of super-reduction.

In the material presented triploid and diploid aberrant plants, obtained after isolation, were distinguished by similarities of certain characteristics as compared with maternal plants. But definite differences in reaction could also be shown, indicating a special influence by the chromosome set obtained, in the first case mainly of a qualitative nature, in the other probably a combination of quantity and quality.

In the case of two of the triploid aberrants obtained after crossing, the chromosome number of the father plant is not known and therefore they will not be treated here. The other two triploid aberrants are all the more interesting in that they constitute a cross between an apomictic strain and the sexual strain 709.

♀		♂			Aberrant Percentage seed setting	
Strain	2n	Strain	2n	2n	Isolation	Free flowering
701	50	709	90	$\pm$ 90	55,8	88,0
703	50	709	90	$\pm$ 95	18,3	75,5

An evident influence by both parent plants on various properties

could be shown in both plants. One of them at least had a greater seed yield than the mother plant. The two aberrants have been tested in F_2 and in particular F_2 progeny of the cross 703 × 709 has been studied and also to a certain extent the F_3 progeny.

O. F_2 AND F_3 OF DIPLOID AND TRIPLOID ABERRANTS.

The F_2 of the aberrant plants described above are of great importance in order to control, among other things, whether the F_1 has an apomictic or a sexual seed formation. It has already been mentioned that the haploids found by MÜNTZING and KIELLANDER seem to be

Scheme 3. The origin of the 8 aberrant plants.

F_1		♀		♂		F_1	F_2 family	
No.	2n ±	Strain	2n	Strain	2n	Type	No.	Origin (is. = isolation sp. = free flowering)
G 186/3	66	702	50	Not known		Dipl. cr.	Ä 76	is.
G 220/1	72	768	86	708	64	» »	Ä 747	sp.
G 193	95	703	50	709	90	Tripl. cr.	Ä 720	is.
Ä 67/3	90	701	50	709	90	» »	Ä 890	is.
G 223/3	75—80	5303	78	5303	78	Dipl. isol.	Ä 750, 751	is., sp.
G 190/6	48	702	50	702	50	» »	Ä 711	is.
G 226/1	110—115	5303	78	5303	78	Tripl. »	Ä 754	sp.
G 186/1	74	702	50	702?	50	» » ?	Ä 74	is.

sexual, and that also appears to be true, at least frequently, of the diploid and triploid aberrants (ÅKERBERG, 1939; MÜNTZING, 1940). This was ascertained in our own material by the F_2 being variable in morphology and chromosome number, while MÜNTZING found it in determinations of the chromosome numbers of the F_2 of triploid twin aberrants. MÜNTZING, however, also found certain plants in which the deviating number indicated fertilization of an unreduced egg-cell, which may have had an aposporous origin.

Above an account was given of four different types of diploid and triploid aberrants, and below we shall deal with the F_2 and F_3 of 8 satisfactorily controlled plants, having two representatives in each of the four groups. The data for the F_3 are for the present only preliminary. The origin of the 8 plants is shown in Scheme 3.

The first F_1 plant, G 186/3, was obtained from strain 702 after free

flowering. It has $2n = \pm 66$ and was assumed to be a diploid aberrant, derived after crossing. It may also be assumed to be a self-fertilized triploid aberrant. But as the mother strain, 702, has poor pollen and consequently cross-fertilization must have taken place, the most probable assumption is that the plant is a diploid. With regard to six of the F_1 plants the origin is more certain. The 2n, it is true, has not been determined for G 223/3 and G 226/1, but on the basis of the F_2 results the approximate values recorded may be regarded as fairly reliable. The last F_1 plant, like the first one, originates from strain 702 after free flowering. The chromosome number is suggestive of self-fertilization of an unreduced egg-nucleus, but that is not compatible with the above statement that strain 702 has poor pollen. The question as to the mode of origin must therefore remain open. The F_1 plants also differ from each other in that they originate from mother plants having quite different chromosome numbers.

Germinability and polyembryony in seeds from F_1 plants. — So far the investigations of the germinability of seeds from F_1 plants are not so extensive. In small experiments seeds of different ages from 13 F_1 plants (including some of the 8 plants mentioned above) originating from strains 702, 703 and 5303 have been investigated. It was found that the F_1 plants had an average germinability of 73,9 per cent and the maternal plants 82,6 per cent (Diff. $8{,}7 \pm 3{,}57$, $P < 0{,}05$). All the F_1 plants were not, however, inferior to the maternal plants in this respect, some of them showed good germinability, but the variation was much greater in the former (52—96 per cent as compared with 75—87 per cent). These experiments suggest that some of the F_1 plants, as in the case of the sexual strain 709, have a lower seed germinability than the apomictic plants.

In the germination experiments a study was also made of the occurrence of polyembryony. A rather extensive material of the four F_1 plants, G 186/3, G 193, G 223/3 and G 226/1 as well as a number of plants of the F_2 families are available, and they are compared below with the mother plants, the data for which are taken from Table 13.

There is obviously a distinct difference in the frequency of polyembryony in these F_1 plants, but the figures do not indicate any connexion with the origin of the plants. Similar differences have also been obtained in investigations of other F_1 plants. In the F_2 one of the families, G 226/1, shows a slight tendency to an increase of polyembryony. (χ^2 after correction of continuity 8,71, $P < 0{,}01$). Otherwise the F_2 plants agree with the corresponding F_1 plants.

TABLE 21. *The frequency of polyembryony in certain F_1 and F_2 plants compared with the mother plants.*

	Strain			F_1 plants			
	702	703	5303	G 186/3	G 193	G 223/3	G 226/1
Number of germinated seeds	118	100	293	63	272	164	160
Percentage polyembryony ...	7,6	9,0	27,3	0	12	16	0
				F_2 plants			
Number of germinated seeds				76	100	48	120
Percentage polyembryony ...				1	9	15	7

Morphology, chromosome number, etc. in F_2 and F_3. — From the eight F_1 plants nine F_2 families were raised, six of them after isolation and three after free flowering. The F_2 families show agreement in certain respects, but differ distinctly in others. Measurements of two characters, height and tuft circumference, have been made in all plants in certain families. The values obtained are shown in Tables 22 and

TABLE 22. *Height and circumference of tufts (spreading capacity) in the F_2 families Ä 76 and Ä 751 compared with the maternal plants (strains 702 and 5303 respectively).*

Strain or	Height in cm.												Number of
Family	< 40	45	50	55	60	65	70	75	80	85	90	95	plants
702							5	4	6	1			16
Ä 76		2		1	1	4	2	5	2	1			18
5303							1	2	6	1			10
Ä 751							1	1	4	4	5		15[1]

Strain or	Tuft circumference								Number of
Family	< 40	60	80	100	120	140	160	180	plants
702				2	16	13	1		32
Ä 76	1	1	8	6	2				18
5303					5	9			14
Ä 751		1	1	5	6	2	2		17

23. Table 22 is an example of the nature of the variability in two F_2 families, Ä 76 and Ä 751, both from diploid aberrants. In all families except Ä 720 the variation was about the same as in the example given. The comparison made between σ^2 for F_2 families and maternal plants will therefore be an expression of the segregation in the respective

[1] A further two plants without panicles.

property. It will at once be seen that the segregation is of a very complicated nature if sexual seed formation takes place. In all families the variability in at least one of the characters was greater ($P < 0{,}01$) than in the maternal plants. Thus all of them showed evident segregation, indicating at least partly sexual seed formation in the F_1 plants. Family Ä 720, however, gave somewhat different results in these characters. The great variation found in this family is due to the presence of a few

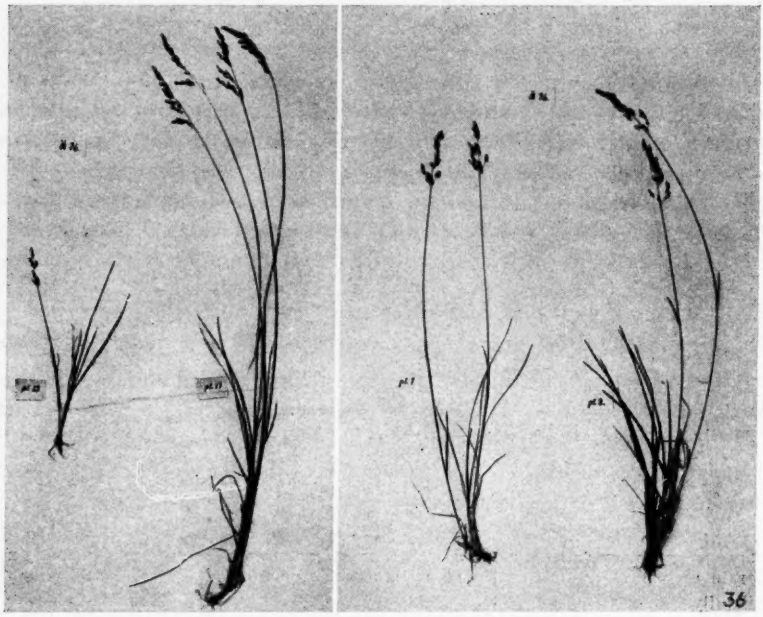


Fig. 36. Four plants from the F_2 family Ä 76.

widely divergent plants (possibly haploids; cf. below). Other plants showed less variation. The family therefore did not exhibit a similar segregation to that of other families, and as far as height and spreading capacity are concerned it cannot be so plainly distinguished from, for instance, a family with mostly maternal plants and only solitary aberrants. That the family is certainly variable, however, will be shown below.

The F_2 family Ä 76, obtained after isolation of G 186/3, seems to be one of the most variable families and at the same time the weakest vegetatively. By way of comparison it may be mentioned that in a small yield test Ä 76 was found to be

TABLE 23. *Comparison between F_2 families and maternal plants with respect to height and circumference of tuft (spreading capacity). Variability and average values.*

Strain or Family	Height in cm.							Circumference of tuft in cm.						
	n	M	σ^2	$\frac{\sigma^2}{\sigma^2}$	P	Quotient $\frac{\text{inter}}{\text{intra}}$	P	n	M	σ^2	$\frac{\sigma^2}{\sigma^2}$	P	Quotient $\frac{\text{inter}}{\text{intra}}$	P
702	16	78,6	22,22	—	—	—	—	32	139,1	195,79	—	—	—	—
Ä 74 F_2	18	65,9	118,69	5,34	$<0,01$	18,70	$<0,001$	18	90,8	911,06	4,65	$<0,001$	22,48	$<0,001$
Ä 76 F_2	18	59,8	139,47	6,28	$<0,001$	35,60	$<0,001$	18	81,2	386,29	1,97	$=0,05$	146,71	$<0,001$
Ä 711 F_2	6	75,7	102,36	4,61	$<0,01$	0,91	$>0,2$	6	111,3	218,68	1,12	$>0,2$	19,62	$<0,001$
703	6	81,8	3,76	—	—	—	—	11	137,5	201.36	—	—	—	—
Ä 720 F_2	18	68,2	358,03 [1]	95,22	$(<0,001)$	3,03	$<0,2$	18	111,2	1,312,76 [1]	6,52	$(<0,01)$	5,21	$<0,05$
5303	10	81,3	17,34	—	—	—	—	14	145,2	97,98	—	—	—	—
Ä 751 F_2	15	86,2	30,76	1,77	$<0,2$	5,64	$<0,05$	17	106,1	588,43	6,01	$<0,01$	31,82	$<0,001$
Ä 754 F_2	18	57,3	34,35	1,98	$<0,2$	129,75	$<0,001$	18	116,7	394,12	4,02	$<0,01$	24,03	$<0,001$
768	13	65,8	14,31	—	—	—	—	15	125,7	393,67	—	—	—	—
Ä 747 F_2	24	60,9	76,43	5,34	$<0,01$	3,70	$<0,2$	24	112,4	475,20	1,21	$>0,2$	3,67	$<0,2$
701	4	54,8	4,92	—	—	—	—	—	—	—	—	—	—	—
Ä 890 F_2	12	42,5	79,73	16,21	$<0,01$	7,05	$<0,05$	—	—	—	—	—	—	—

[1] See text.

much inferior to strain 702 in yield. In this family it was quite impossible to refer a number of the plants to the same type. No two plants were exactly alike. In addition to the variation in height and in tuft circumference variation was also observed in leaf-length, leaf-breadth, colour, shape of panicle, etc. The chromosome numbers were determined in three plants and were found to be $\pm$ 59, $\pm$ 70 and $\pm$ 93 respectively. The last-mentioned seems to have arisen from an unreduced egg-cell. Two plants have been tested in F_3. In one of the F_3 families an evident morphological variation was observed.

Even the other family Ä 747, of the same type as Ä 76 but obtained after free flowering of the F_1, was so variable that no two plants could be said to be alike, but they nervertheless belonged to a similar type. An intense segregation was shown, among other characters, in the anthocyanin pigment (cf. p. 56). The family was not, however, vegetatively weaker than the mother plant. The chromosome number was investigated in three twin pairs, and was found to be (1) $\pm$ 76 and $\pm$ 110, (2) $\pm$ 87 and $\pm$ 110 and (3) $\pm$ 73 and $\pm$ 74. In cases 1 and 2 it is probable that one of the twins arose from an unreduced (aposporous) egg-cell. Case 2 constitutes a new instance in which both twins have certainly a different chromosome number from that of the mother plant ($2n \pm 72$), one of the twins, as expected, originating from an unreduced egg-cell (cf. p. 50). In this case both twins were also highly differentiated morphologically.

The F_2 family Ä 720, obtained after isolation of G 193, did not show, as already mentioned, the same segregation in height and tuft circumference as the other families, but a positive agreement with the latter could be ascertained as regards other morphological characters. The majority of the plants had several characteristics reminiscent of the father plant 709 (e. g. panicle more open and with few spikes), while a few of them showed a panicle more in agreement with that of the mother plant 703. Most plants in the family were not pruinous. We find in this family a similar segregation to that previously described in the cross 709 $\times$ 702 (p. 46). Three plants were much weaker than the others (haploids?). Seed was obtained from only one of these plants. Ä 720 is somewhat vegetatively weaker than 703. In a small yield test with two harvestings the decrease in yield was more than 20 per cent. Chromosome counts were made only in one plant. It had $2n \pm 86$. Two plants have been tested in the F_3. The F_3 families differed distinctly from each other morphologically.

The other F_2 family of the same type is Ä 890. Ä 890 was obtained after isolation of the F_1. It was planted out under conditions that did not afford the same facilities for observing the morphological characters as in the other F_2 families described here. The plants were of about the same type, but an evident variation in height could be observed. Only one morphologically main type could be distinguished. The number of plants, however, was lower than in Ä 720. Two F_2 families, planted out at the same time as Ä 890, also originating from aberrant plants of the cross 701 $\times$ 709, showed a very great morphological variation. One family had extremely broad-leafed types. Unfortunately the chromosome numbers of the F_1 of these families were not counted.

The F_2 families Ä 750 and Ä 751 were obtained from G 223/3, the former by isolation, the latter after free flowering. As the former family consists of only one plant it will not be treated separately. An evident variation was observed in panicle formation, presence of anthocyanin, colour, etc. It was possible, however, to refer

many plants to a certain type having several characters in common. Only one such type was obtained. This family also contained a few extreme plants that did not form panicles. This family is of special interest, however, owing to the fact that it is somewhat taller than the mother plant 5303 (the F_1 was almost as tall as 5303). That observation was made only in this F_2 family. In spreading capacity, however, it is inferior to the mother plant, and in a yield test it was found to have about 20 per cent less yield. The chromosome numbers of five plants were $\pm$ 77, $\pm$ 74, $\pm$ 76, $\pm$ 109 and $>$ 100 respectively. In the two last-mentioned plants an unreduced egg-cell seems to have been fertilized. The first three numbers have justified the assumption of the chromosome number of 75—80 for the F_1.

Family Ä 711, obtained after isolation of G 190/6, consisted of only six plants. This family did not differ much from the mother plant 702 in its morphological characters, but a positive difference could nevertheless be detected, among others, in its spreading capacity. The family was variable, which was apparent, for instance, in the height of the plants. Two of the plants could not be distinguished from each other for certain. The chromosome numbers of three of the plants were $\pm$ 46, $\pm$ 48 and $\pm$ 72 respectively. In the last-mentioned case we have another instance of the fertilization of an unreduced egg-nucleus.

Ä 754, obtained after free flowering of G 226/1, belongs to the fourth type of F_2 families, where the F_1 is a triploid aberrant arisen after isolation. It is morphologically quite different from Ä 751, it is shorter, has broader leaves and a somewhat better spreading capacity. Its morphological variation is plainly evident, for instance, with respect to spreading capacity, panicle formation, colour and shape of panicle, vegetative leaves, etc., but here, too, several plants appear to belong to a certain type, even if they vary among themselves. Vegetatively, the family is weaker than the mother plant. The chromosome numbers of three plants were $\pm$ 114, $\pm$ 112 and $\pm$ 114 respectively, and the assumption of $2n = 110$—115 for the F_1 is based on these numbers.

The last F_2 family, Ä 74, concerning which there is a great uncertainty as to the derivation of the F_1, was obtained by isolation of the F_1 plant. It is characterized by an evident morphological variation. It was nevertheless possible to differentiate a certain morphological type, to which a small number of the plants could be referred. As regards some properties, tillering and panicle shape, the plants seemed to be more typical *angustifolia* than the mother strain 702. The family was vegetatively weak, but not quite so weak as Ä 76. The chromosome numbers of three plants were $\pm$ 73, $\pm$ 72 and $\pm$ 74 respectively.

Mention was made above that F_2 families had in certain cases been compared with the mother strains with respect to yield. In the test made altogether five F_2 families were compared with their respective mother strains. About 50 plants of each family and strain were grown in an area of 1,5 square metres. In this test two harvestings were made in one and the same year. The yield of the F_2 families in this test was on an average 20 per cent lower than that of the mother strains. Only the F_2 family Ä 747 mentioned above attained the same yield as its mother strain.

Some of the plants of the F_2 families Ä 76, Ä 720, Ä 751 and Ä 754 have been examined as to germinability. The results cannot be regarded as the average figures for the families, as plants having a sufficient number of seeds had to be selected. The germinating capacity of the F_2 plants is compared below (Scheme 4) with that of the mother plants investigated on the same occasion. All seeds were two years old.

From the results of this comparison it is clear that there is a certain difference in germinability between different plants of the same family and also between different families. On account of the origin of the material the latter difference is not of any great interest, but a more interesting feature is, as appears from the figures, that even in the F_2

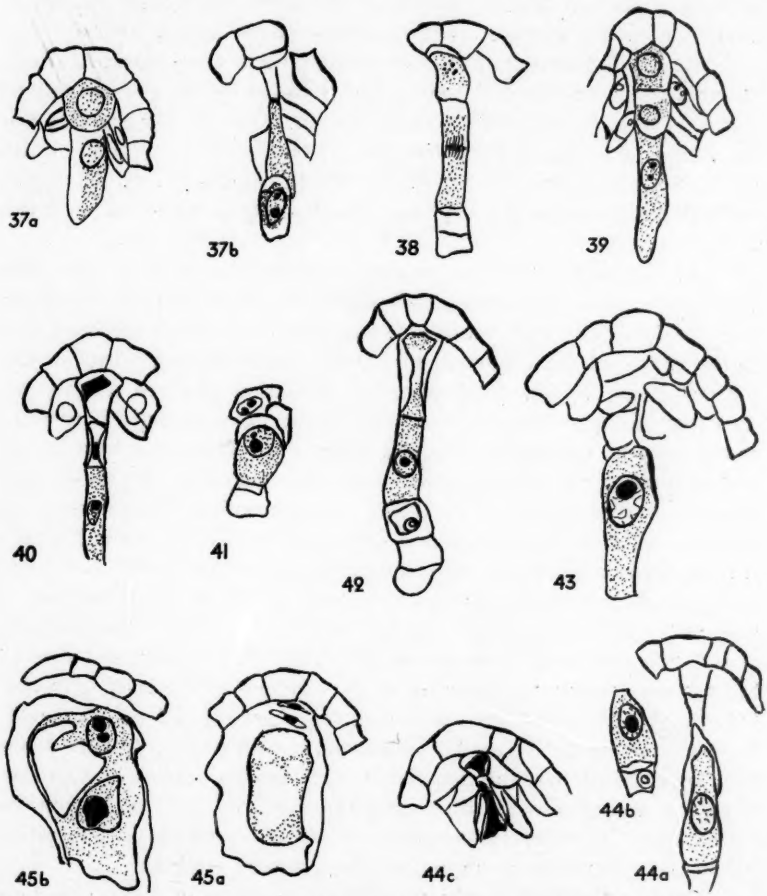
Scheme 4. Germinating capacity.

Mother strain		F_2 plants				
No.	% germination	Family No.	Percentage germination			
			1	2	3	4
702	76	Ä 76	72	50	83	—
703	75	Ä 720	78	76	77	—
5303	75	Ä 751	70	68	—	—
—	—	Ä 754	58	60	46	76

plants occur with a lower capacity of germination than the mother strains.

Formation of male and female gametophytes. — The study of the formation of the male gametophyte was confined to investigations of stamens and pollen. In many cases great disturbances were observed in the quantitative and qualitative pollen formation, indicating different combinations of diplontic and haplontic sterility. Out of 34 plants belonging to 7 of the F_2 families discussed above (all except family Ä 890) 17 had a distinctly inferior stamen formation (in the shape of shrivelled or undehisced stamens) than the mother strains, and of 36 investigated plants 11 had inferior pollen (inferior in both cases denotes a difference of > 20 per cent). The pollen formation in several cases was practically nil. Plants with both impaired stamens and poor pollen were obtained in *all* families. In family Ä 720 the reduced pollen formation was restricted to the weak plants, but in the other families defective stamens and pollen were found even in plants which, vegetatively at least, were up to the average. This was true of, among others, the vegetatively vigorous family Ä 747. It is not possible from the material

to determine whether the families show different degrees of variation with respect to pollen formation. It is evident, however, that as far as



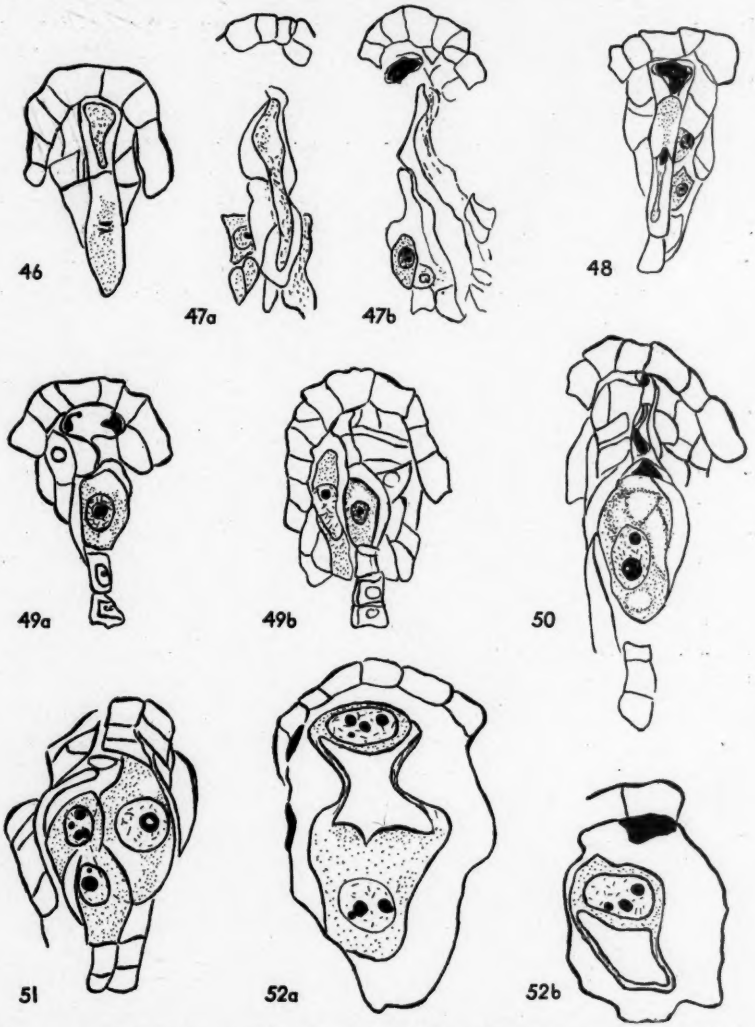
Figs. 37—45. The embryo-sac development in two aberrants, Ä 67/3 and G 193 (*a* and *b* adjacent sections from the same nucellus). — Figs. 37—41. Ä 67/3. — 37 *a*. The dyad. — 37 *b*. One of the dyad cells and an aposporous E.S. initial. — 38. Second division, most of the chromosomes in the micropylar cell are in another section, division here not quite normal. — 39 and 40. Three megaspores, in 40 two of them degenerated. — 41. The developing megaspore. — Figs. 42—45. G 193. — 42. Three megaspores, two of them degenerated. — 43. The developing megaspore. — 44 *a*—*c*. The developing megaspore (*a* and *b*) and two degenerated ones (*c*). — 45 *a* and *b*. An embryo-sac with two of the nuclei in *b* and the degenerating megaspores in *a*.

this property is concerned there is a marked segregation within the families. Thus such a segregation characterizes all families irrespective of their mode of origin. Further, the pollen formation of the F_2 families is on an average lower than that of the mother strain.

The development of the female gametophyte was studied in two of the previously described F_1 plants Ä 67/3 and G 193 and in a couple of plants of the F_2 of G 193 (family Ä 720). The two F_1 plants had the following derivation: Ä 67/3 $(2n = \pm 90) = 701$ $(2n = 50) \times 709$ $(2n = 90)$, and G 193 $(2n = \pm 95) = 703$ $(2n = 50) \times 709$. Thus both are triploid aberrants of an apomictic mother strain and a sexual father strain.

In both the F_1 plants the megaspore formation seems to take place along the same lines as already described for strain 709 (see p. 18 and Figs. 5—11). The material does not permit of a more detailed analysis of the course of the reduction division. It was evident, however, that the reduction division resulted in the formation of three megaspores. As a rule the upper dyad cell does not divide. Occasionally four megaspores may be formed (v. Fig. 38, where both dyad cells were at second anaphase). In all controlled cases the embryo-sac developed from the chalazal megaspore. In G 193 no differentiated aposporous cell could be detected in the examined sections. The E.S. shown in Fig. 45 *a* (the nuclei in adjacent sections) probably also originates from a megaspore, judging from its position in relation to the degenerated rests of the other two megaspores. In one case in Ä 67/3 an unmistakable aposporous cell could be observed (Fig. 37). But in this case it seems to have developed late, compared to the products of the E.M.C. In both G 193 and Ä 67/3, however, numerous twins are formed (cf. Table 21). In order to explain the sexual seed formation in the F_1, it might therefore be assumed that the aposporous E.S. initials appear late and are unable to compete with the legitimate E.S. In this connexion the rather high frequency of triploids in the F_2 progeny is interesting. Among 24 controlled plants no less than 6 were triploids.

The two embryologically investigated F_2 plants of Ä 720 (the F_2 of G 193) were also examined with regard to the formation of stamens and pollen and also seed setting. One of the plants, Ä 720/3, had completely shrivelled stamens, no pollen was obtained and the seed setting was nil in both isolation and free flowering. Moreover, it was vegetatively very weak (haploid ?; see above). The embryological examination gave somewhat different pictures. Fig. 46 shows a second anaphase in the chalazal dyad cell, while the micropylar cell is in process of



Figs. 46—52. The embryo-sac development in two F_2 plants from the aberrant G 193 (*a* and *b* as in Figs. 37—45). — Figs. 46—48. Å 720/9. — 46. The dyad, the micropylar cell degenerating. — 47 *a* and *b*. A degenerating dyad, alongside an aposporous E.S. initial. — 48. Three degenerating megaspores. — 49 *a* and *b*. The developing megaspore, in *a* degenerating megaspore, in *b* an aposporous E.S. initial. — Figs. 50—52. Å 720/3. — 50. A developing megaspore and 2 (3?) degenerating megaspores. — 51. A developing megaspore (to the right) and two aposporous E.S. initials. — 52 *a* and *b*. An embryo-sac in four nucleate stage, three of the nuclei drawn, in *b* rests of degenerating megaspores.

degenerating. Fig. 47 was rather difficult to interpret. The entire nucellus appears here to have degenerated. Two cells, probably dyad cells, were seen in the nucellus, these two cells being also in process of degenerating. Alongside the lower dyad cell there was a nucellar cell closely resembling an aposporous E.S. initial. Two such cells are also seen in Fig. 48, where three megaspores had been formed, all of them degenerated. Finally, in Fig. 49 a well-developed megaspore and also an obvious E.S. initial are seen in *a*. As seed formation very seldom takes place, it is probable that in such a case as that last-mentioned degeneration sets in at a later stage of the E.S. development. The embryological investigation indicates that a degeneration of the embryo-sac formation may take place at different stages and that such a degeneration occurs in spite of the presence of aposporous E.S. initials (diplontic sterility), but haplontic sterility may also occur.

The other F_2 plant, Ä 720/9, was vegetatively vigorous, had good stamen and pollen formation (97 per cent good pollen) and a seed setting of 4,2 and 71,5 per cent after isolation and free flowering respectively. In this plant the megaspore formation seems to take place without any disturbances. Three megaspores are formed as a rule. In Fig. 50 is not sure whether there is not a fourth megaspore. The degeneration has advanced too far to permit us to express a definite opinion. Aposporous cells were clearly observed in certain sections, but not in all. Such a case is illustrated in Fig. 51, showing a well-developed megaspore and two plain aposporous E.S. initials. What will be the result here, an aposporous embryo, a legitimate embryo or a twin, cannot be determined. In two cases an E.S. with four nuclei was studied. It was difficult to determine the origin of the E.S. for certain, but one of the cases at least points to its being legitimate. Ä 720/9 has not yet been examined in F_3, but judging from what has been said above it should at least have partly sexual propagation.

Seed setting. — For the seven F_2 families in which the pollen formation was studied, seed setting in a number of plants selected at random has also been controlled partly in isolation, partly in free flowering. The isolations were made with two isolations per plant, therefore the values obtained represent fairly reliable measurements of the isolation fertility under prevailing conditions (cf. p. 32). Seed setting is always the result of an interaction of several factors, and it is a difficult matter to analyse the combined action of the factors on the different F_2 plants. Among others, we have here the influence of diplontic and haplontic sterility, the degree of sexuality, pollen supply and pollen quality. The

occurrence of apomictic seed formation does not imply that male sterility is always accompanied by female sterility in free flowering.

The seed setting in the investigated plants is shown in Table 24. It has been compared only with the seed setting of the mother strains, as unfortunately neither the father plants nor the F_1 plants were examined in the same year as the F_2 families. The material is not sufficiently extensive in all families for a reliable determination of the variability in seed setting in the F_2 as compared with the maternal plants. In one or two cases, however, there was a suggestion of in-

TABLE 24. *Seed setting in F_2 families compared with mother strains.*

F_2 family	Percentage seed setting								Mother strain	
	0	5	10	30	50	70	90	Average	No.	% seed
Ä 74 isolation	6	1	1					3,5	702	2,7
free flowering	1		4		3			29,7		54,7
Ä 76 isolation	6	1	2					5,6	702	2,7
free flowering	1	1	2	3	2			32,1		54,7
Ä 711 isolation	3							0,9	702	2,7
free flowering				1	2			51,8		54,7
Ä 720 isolation	6	2						2,4	703	4,3
free flowering	2		1	1	2	2		41,4		34,2
Ä 751 isolation	1	1		2				21,0	5303	15,4
free flowering				1	1	2		61,0		67,3
Ä 754 isolation	1		2	1				14,1	5303	15,4
free flowering				2	5	2		61,0		67,3
Ä 747 isolation	5		1					2,0	768	35,5
free flowering			1	4		1		46,6		90,8

creased variation both after isolation and after free flowering. In Table 24 the difference in seed setting between the average of the F_2 plants and the maternal plants is occasionally very considerable. In an analysis of the variation a difference, where P was $\leq$ 0,05, was obtained for the following F_2 families: 1) Ä 76 free flowering (P$<$ 0,05), 2) Ä 74 free flowering (P approximately 0,05), 3) Ä 747 isolation (P $<$ 0,05) and 4) Ä 747 free flowering (P$<$ 0,01). In all these cases the F_2 family had a *lower* seed yield than the maternal plants; in no case did the F_2 family have a demonstrably higher yield of seeds.

Four of the families are derived from the strains 702 and 703, which were characterized by a *poor seed setting in isolation.* The corresponding F_1 did not show a positive difference in seed setting after

isolation from that of 702 and 703, and that also seems to be true of the F_2. In the F_2 family Ä 720 the father plant 709, however, had a good seed setting in isolation. But it should be borne in mind that the same factors do not influence the seed setting in the F_1 and the F_2 as in the parents (provided the latter are apomictic). The F_2 families from 5303 did not on the average show a lower seed setting in isolation than 5303 itself (the F_1 in both cases was considerably weaker), but in both families plants occurred with a very low seed yield, < 10 per cent, and all these plants had a very poor pollen formation. Finally, Ä 747, like the F_1, was a very poor seed producer in isolation, and this weakness was also associated with poor pollen formation. Even if 1938, when these investigations were carried out, was characterized by low seed yields in the isolations (see Table 6), it is nevertheless noteworthy that nearly 80 per cent of the investigated F_2 plants had less than 10 per cent seed yield.

The seed setting in *free flowering* is more interesting than after isolation, inasmuch as in the former it is possible to indicate the causes of the variation somewhat clearly. In three of the F_2 families, Ä 74, Ä 76 and Ä 747, several plants show a considerably inferior seed setting to that of the mother strains, while the other four families do not exhibit any significant difference. One of these four families, Ä 720, has even a higher seed yield than the mother strain, but the value for strain 703 is based on only two plants, one of which had an abnormally low seed yield. Seed setting in the entire F_2 material varies from nil to 82 per cent at most. It is difficult to fix a limit below which the seed setting of an F_2 plant cannot be considered to be in conformity with the seed setting in normally apomictic strains. On the basis of our knowledge of the seed setting and its variation in the apomictic strains in 1938, it may be said with a high degree of probability that the limit of 30 per cent is not too high. In that event 13 of the 47 plants investigated in F_2, i. e. at least 25 per cent, had a demonstrably lower seed yield than the apomictic strains. In the latter none of the investigated plants produced less than 30 per cent seed.

In the 13 plants showing a decreased seed yield the pollen formation was defective in some way or other. Thus 5 of these plants had more or less shrivelled stamens. But there were also plants with shrivelled stamens which produced more than 30 per cent seed. In the entire F_2 material 8 such plants were met with. Among them there were quite extreme cases, for instance, with 100 per cent more or less shrivelled stamens and more than 70 per cent seeds. As the pollen

grains obtained in these cases were morphologically good, diplontic sterility must be assumed as the probable cause of the degeneration of the stamens. That seed formation despite such an extreme stamen degeneration can take place quite normally has been shown in apomictic strains, among which analogous examples have been recorded. With regard to the F_2 plants in question the most plausible assumption seems to be that they are apomictic or that diplontic sterility has only influenced the formation of stamens. As this was also controlled in the pollen investigations it is less probable that in the example cited we have to do with only a modificatory change in the stamens from the *examined* panicle.

Whenever the pollen formation showed no great defect, either quantitatively or qualitatively, the seed setting was always above 30 per cent. But a satisfactory seed setting was found associated not only with a poor stamen formation but also with an inferior quality of pollen. The latter, however, is not so remarkable as the former. Poor seed setting in free flowering was as a rule associated with vegetative weakness. But that has not, however, always been the case.

Summary. — Judging from the aberrant plants investigated so far — irrespective of whether they were haploids, diploids or triploids of one kind or other — a plant of an apomictic *Poa pratensis* arisen by fertilization appears to acquire sexual seed formation. Nothing definitely can be said as to whether there are any gradations of the sexuality thus occurring. Experiences from investigations of polyembryony indicate, however, that certain F_1 plants possess a greater tendency to apospory than others. The occurrence of apospory in the F_1 also seems to be probable, partly from plants in the F_2 with a chromosome number suggestive of the fertilization of an unreduced egg-cell, partly from the results of the embryological investigations. The material submitted here does not furnish a reliable picture of the seed formation of the F_2 plants. Some of them have shown an evident morphological variation in the F_3.

An especially interesting question is how the aberrant plants and their offspring may be expected to behave in a natural selection. The formation of aberrants frequently seems to involve a disturbance also in vitality and fertility, and several of the aberrants were from the zygote stage right up to the complete seed-producing plant inferior in various characters to the apomictically originated plant. The inferiority is of such a nature that the occurrence of aberrants is proportionately less in nature than that obtained in these experiments. The

capacity of the aberrants to produce offspring also appears to be frequently decreased. It should also be pointed out, however, that in certain cases the aberrants were clearly superior to the mother strains in vitality or fertility. There is a tendency that such aberrants occur preponderatingly among those arising after crossing.

In the above account a description was given of changes in certain characteristics in F_2 families as compared with the mother strains. No comparison with the father strains was as a rule possible. In several cases, however, the mother and father strains were the same. An attempt is made in Table 25 to summarize the results of com-

TABLE 25. *Summarizing comparison between F_2 families and mother strains.* (*See text.*)

F_2 family	Germination F_1	Germination F_2	Height	Circumference of tuft	Yield	Seed setting in free flowering
Ä 76		±	−	−	−	−
Ä 747			±	±	±	−
Ä 720	±	±	±	−	−	±
Ä 890			−			
Ä 751	−	±	+	−	−	±
Ä 711			±	−	−	±
Ä 754	±	−	−	−		±
Ä 74			−	−	−	−[1]

parisons between the various characteristics, but owing to the scarcity of the material there is a slight uncertainty in certain respects. The F_2 families have been designated by +, ± and −, according to whether they were (1) taller, larger, (2) equal to, or (3) shorter, smaller than the mother plants. A difference has been recorded only when the probability of such a difference was $\geq$ 0,95. Of the properties recorded, height and spreading capacity need not of course signify a minus in the natural selection. Under certain habitat conditions *Poa pratensis* is short, in other habitats it has a weak formation of extra-vaginal shoots. But as far as the F_2 family discussed here is concerned the spreading capacity at least is in all probability associated with the vitality, for in these families it was not observed that an inferior spreading capacity was accompanied by an increased formation of intra-vaginal shoots. The two families Ä 74 and Ä 76 were especially controlled in this respect. It is also conceivable that the inferior

[1] P approximately 0,95.

spreading capacity and the lower yield in the F_2 families of *triploid* aberrants were also due to a slower development in these families. But the material submitted does not lend support to such an assumption. The two families Ä 711 and Ä 74 of strain 702 were diploid and triploid respectively, and families Ä 751 and Ä 754 derived from strain 5303 were respectively diploid and triploid. In the former case the diploid family has a superior spreading capacity, but in the latter case the triploid is the better. It is therefore probable that a lower yield and a slower spreading capacity in the *triploid aberrants* are also the results of a lower vitality.

A + occurs in only one case, for family Ä 751, which was on an average taller than the mother strain, from which it was obtained by isolation. The occurrence of ± and — is very variable in the families. As an instance of this variation we may mention families Ä 747 and Ä 74, the former having the symbols ±, ±, ± and — for height, spreading capacity, yield and seed setting, while the latter shows the symbols —, —, — and — for the same properties, seed setting showing no particularly marked difference. All families show at least a minus for some character undoubtedly connected with vitality or fertility. This characteristic but heterogeneous deterioration shown by the F_2 families described is of course partly due to the properties of the F_1 plant, partly to its acquired sexuality. As shown above, in sexual seed formation an intense segregation occurs, many plants becoming less vital or less fertile and consequently the F_2 on the average shows a deterioration. In this respect these F_2 families very closely resemble the progeny of the sexual strains 709 and 813 (see p. 41).

The less vital and less fertile plants will undoubtedly be eliminated by natural selection, but the F_2 also contained plants that were in various respects at least as good as the mother strains, and among them there may also be an occasional plant superior to its mother strain in some particular property, e. g. height in family Ä 751. A tendency to apomictic seed formation was also observed in the F_2 families, probably to a varying extent in different families. This tendency to apomictic seed development was shown by embryological studies, by polyembryony and by the occurrence of unreduced egg-cells. The F_3 material studied so far does not furnish an answer to the question as to whether already in the F_2 any plant had a predominantly apomictic seed setting. It should be expected, however, that among those plants remaining in nature, there should also arise, as the products of segregation, in a subsequent generation of the F_2

families described here valuable new strains, from the standpoint of selection, with mainly apomictic seed setting. The progenies of these strains will then be for the most part constant with the selective value of the mother plant. It should, however, also be possible for similar strains to arise possessing a predominantly sexual seed formation. But the experimentally produced material furnishes no definitive answer to the question whether there is among them some strain in which the reduction division is so little disturbed that the following generation will not show any great segregation, or whether there is possibly any connexion between, for instance, vitality and heterozygosity. A more detailed discussion of these important questions, i. e. the transition from sexuality to apomixis and from apomixis to sexuality must therefore be put off until the data for the collected material are presented.

2. PROGENY INVESTIGATIONS OF MATERIAL COLLECTED IN NATURE.

A. INTRODUCTORY NOTES.

Of the 12 strains treated in the foregoing pages 10 were thus mainly apomictic, while two, both belonging to the form complex *irrigata*, had predominantly sexual seed setting. The latter two strains originate from one and the same population. The investigations of the progenies of the apomictic and sexual strains lead to the assumption that sexual types from apomictic strains (by aberrant formation) as well as apomictic types from sexual strains (by segregation) may arise in nature. Thus we have here to a certain extent a reversible process.

In the majority of cases the investigations of apomictic seed setting in *Poa pratensis* were made on a relatively small number of individuals. In these investigations (MÜNTZING, 1933 a; RANCKEN, 1934; FLOVIK, 1938; KIELLANDER, 1941 a, and others) solely apomictic types were also met with. An investigation of the seed production of a large number of types, mostly American, was made by TINNEY and AAMODT in 1940. The seed setting was controlled only by means of progeny examinations (cf. below). Altogether 11,696 plants of 102 families were planted out. Of these families 48 were morphologically constant, while the other 54 families showed an aberrant frequency varying between 0,44 and 21,92 per cent. The average frequency of aberrants for the entire material amounted to only 1,59 per cent, thus a much lower frequency than that obtained in the 12 strains described above (Table 17). The cause of this low frequency of aberrants may of

course be due to the origin of the material, but may also be due to the method of seed germination. The seeds were sown in small dishes, where the seedlings consequently grew up in competition with others. It is therefore probable that the weakest aberrants succumbed already during germination. The aberrants were not investigated cytologically, but were assumed to be both diploid and triploid or possibly mutants. No investigation of the seed formation was made.

Thus TINNEY and AAMODT did not show any purely sexual types but only partly sexual ones. This may possibly be connected with the origin of the material and the habitats from which they were collected. Judging from the frequency of aberrants, the occurrence of purely sexual strains should also be exceedingly low, even if all aberrants become highly sexual.

Our own material for a similar investigation, described here, was obtained partly by collecting panicles from plants in nature (it was frequently impossible to determine whether these plants were wild or only adventitious), partly by procuring samples of seeds. In the latter case the plants were first grown in culture, from which progenies were then raised. The two categories will be treated separately below.

B. THE OCCURRENCE OF APOMICTIC AND SEXUAL SEED FORMATION.

With our present experience from cytological, embryological and genetic investigations of *Poa pratensis* it may be said that the assumption of a predominantly apomictic seed formation in the mother plant from a morphological constancy in its progeny is very rarely erroneous. It seems to be extremely improbable that a morphologically uniform progeny can be obtained from a self-fertilized sexual *P. pratensis* of such a type as that described in the preceding pages. Thus if morphological constancy renders apomictic seed setting probable, then we cannot on the other hand form any definite opinion from the frequency of aberrants as to the degree and type of sexual seed production. For that an investigation of the aberrants is required to ascertain whether they are triploids, diploids or haploids. As mentioned above, the progeny test as a measure of the type of seed development was employed by TINNEY and AAMODT (1940) and also by the present author in an earlier published paper (ÅKERBERG, 1939). In the latter paper a brief report was given of the results obtained from part of the material collected in nature. This material now comprises types from Sweden, Norway, Denmark, Finland, Germany and England, and

the morphological analysis has to a great extent been supplemented with chromosome counts. By this means previous assumptions of apomictic and sexual seed formation have in a high degree been confirmed.

The seeds from panicles or seed samples were germinated in the manner already described (p. 46), the seedlings planted in pots, fixed and afterwards planted out in the open. From 5—10 plants of the majority of the families, of others considerably more, were planted out. Thus the number of plants per family was as a rule not high, and consequently does not permit of any grading of the apomictic seed formation in the respective families, being employed only to distinguish intensive apomictic seed development and intensive sexuality. That such a division is justified in the material in question will be shown by the following survey.

Altogether 96 families have been investigated. In six of them the number of plants was so small that nothing can be said of their seed setting. They are recorded only in connexion with the survey of all chromosome numbers found. In 9 families the morphological variation was so great that no two plants could be said to be alike. In the case of two families, one from Denmark, the other from Germany, the analysis of the plants was very difficult. A rather large plant material of both families was available, and the records and certain crosses seem to indicate that they are predominantly sexual but with a relatively slight morphological variation. Further investigations, however, are necessary before the seed formation of these two types can be established for certain. Their chromosome numbers are recorded, followed by a note of interrogation, in Table 26 among the sexual types.

Of the other 79 families 559 plants were planted out, 22 of which, or 3,9 per cent, were classified as indubitable aberrants, while 5, or 0,9 per cent, showed no distinct divergence. In order to study the distribution of these sure aberrants an attempt has been made to compute the frequency of aberrants among 5 plants, i. e. the lower frequency for the number of plants per family. The following values were obtained:

	Number of aberrants per 5 plants			
	0	≤ 1	2	3
Number of families.....	63	12	4	0

The method gives a somewhat higher frequency of aberrants than that actually obtained, 5,1 as against 3,9 per cent. It is clear, however, that there is no smooth transition to those families in which all plants

differed from each other. Such a classification as that suggested above therefore seems to be possible. The aberrant frequency in these 79 families is lower than that obtained for the 12 families previously treated. This difference is probably an expression of a different formation of aberrants in the investigated strains. But the two collections of individuals are for various reasons not quite comparable for such an analysis. On the other hand, the frequency of aberrants is higher than that in the material investigated by TINNEY and AAMODT. Moreover, families occurred with a high degree of sexual seed formation and in a strikingly high frequency, higher than might have been expected from the frequency of aberrants found. It is extremely difficult to determine what may have been the cause of this. It is possible that the value for the aberrant frequency was too low owing to the small number of plants per family.

Finally, it should also be pointed out that the material does not exclude the possibility of the occurrence of *all* degrees of apomictic seed formation. As mentioned above, it was merely impossible to make such a gradation owing to the small number of plants per family. It appears, however, as if certain combinations of apomictic and sexual seed formation are less frequent.

C. MORPHOLOGY, CHROMOSOME NUMBER, ETC. OF THE FAMILIES SHOWING A HIGH DEGREE OF SEXUAL SEED FORMATION.

The two sexual strains 709 and 813 previously described belong to the form complex *irrigata*. Of the nine new families with a high degree of sexual seed formation one was typical *angustifolia*, one closely resembled *irrigata* and the remaining seven belonged to *eu-pratensis*. From this we see that sexual types in different form complexes were obtained in nature. The only form complex of which no sexual type was obtained is *alpigena*. Six of the nine families were collected in Sweden, two originate from Germany and one from Russia.

As a rule the *morphological* variation was very distinct. Records of morphological characters were repeatedly made during two successive years. The plants in one and the same family most frequently belonged to one and the same main type. — A very singular family of this kind was obtained from Ångermanland. In the vegetative stage the tuft most closely resembles the tuft of red fescue with very narrow leaves (2—2,5 mm. in breadth). — Several properties have been measured. The variability of the properties varies with the families. As an

example of the variability we may give the values for height in two sexual and two adjacent apomictic families.

Scheme 5. Variability in height.

Family	Type	Height in cm. for plant No.				
		1	2	3	4	5
Ä 647	sexual	43	62	26	18	72
Ä 648	apomictic	62	53	62	[1]	66
Ä 654	sexual	79	41	44	57	53
Ä 655	apomictic	55	60	54	50	58

For six of the nine families the morphological variation was controlled in the following generation. In all cases an evident variation

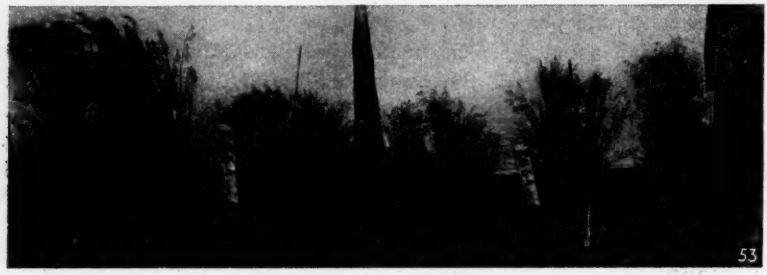


Fig. 53. Family obtained from a sexual plant collected in nature.

was observed between families of the same origin. As an example of this variation we give below the height for the families derived from Ä 647.

Scheme 6. The height for the families derived from Ä 647.

Mother plant		Progeny family	Plant height in cm.
No.	Height in cm.		Average
Ä 647/2	62	Ä 8026	$44{,}5 \pm 1{,}29$
» /3	26	Ä 8027	$30{,}7 \pm 1{,}76$
» /5	72	Ä 8028	$45{,}2 \pm 2{,}33$

Ä 8026 and Ä 8028, which do not differ for certain in height, were clearly distinguishable in other properties. It is not possible to submit

[1] Aberrant, height 46 cm.

any measurements of the vitality of the sexual families as compared with those of the apomictic families. An evident segregation of less vital types occurred in most families, for instance in Ä 647. But some of them were on an average markedly luxuriant vegetatively.

TABLE 26. *Chromosome numbers in material obtained from collected panicles and from acquired seed samples.*

Origin	Apomictic types 2n							Sexual types 2n					
	50	60	70	80	90	100	<130	50	60	70	80	90	100
Material from collected panicles													
Sweden													
Gõtaland	4	1		13	2				3	1	1		
Norrland		5	2	1					1				
Norway, Denmark, Finland	1	3	4						1?				
Germany	6	3	1			1			1	1?			
England				2	3								
Material from acquired seed samples													
All	2	2	1						1				
Seed setting unknown													
Sweden		1	2										
U. S. A.	2		1										
Total	15	15	11	16	5	1			6+1?	1+1?	1		

The *chromosome numbers* of 8 of the 9 families were investigated, and the values are recorded in Table 26, the average numbers for the families being placed in certain classes of chromosome numbers. In six of the families the 2n lies between 60 and 70, in the other two the 2n is higher than 70. The only class in which no sexual types have yet been obtained is 2n = 50—60. The chromosome number was investigated in several plants of the same family. The following variation was found: Family Ä 647 2n = ± 59, ± 60, ± 63; Family Ä 654 2n = = ± 62, ± 66; Family Ä 662 2n = ± 73, ± 74; Family Ä 669 2n = ± 86, ± 86; Family Ä 673 2n = ± 63, ± 64, ± 64 and Family Ä 47 2n = ± 59, ± 69. It is only in the last-mentioned case that the variation was so great that one can speak of an evident difference. Thus in no case was a plant obtained which had arisen by fertilization of an unreduced egg-cell.

Seed setting in isolation and free flowering was investigated in several plants from each of the 9 families. Both characteristics were subject to great variations, fertility after isolation being frequently very

low. In the following table a comparison is made between the seed setting in free flowering of all plants from the 9 sexual families investigated in 1937 and 1938 and the apomictic strains of the collected material controlled in the same years. The values recorded for the apomictic strains are often the average for two plants. The variation between two such plants was very small.

TABLE 27. *Seed setting in free flowering in plants from sexual families compared with apomictic families.*

	Percentage seed setting						Total No.
	0	20	40	60	80	100	of plants
Sexual fam. No. plants	6	5	6	12	8		37
Apom. » » »			2	26	22		50

It is clearly evident from the above table alone that plants from the sexual families showed a considerably greater variation than the apomictic families with regard to seed setting (P for variance quotient for the intra-variation $< 0{,}001$). The difference in seed setting, too, is evident, the apomictic families showing a higher yield (Diff. 21,8 per cent, $P < 0{,}001$). None of the latter had a lower seed yield than 50 per cent and the majority had more than 60 per cent. The sexual families did not behave exactly the same with respect to seed setting. In certain families all the plants examined had good seed setting. In others an evident segregation was observed in the investigated plants. Three of the nine plants of the above-mentioned family Ä 654 had < 10 per cent seed setting and the other > 50 per cent. With regard to seed setting we have in these 9 families fine parallels to the families derived from the aberrants.

It has already been mentioned that a new generation of some of the plants of the families under discussion has been raised. The investigations of this new generation have not yet been concluded. In some cases an evident segregation has been observed, but in most cases the families were surprisingly constant. The latter, however, were all raised from seed obtained after isolation, which may perhaps explain why the morphological variation was so slight. It is also conceivable of course that the progeny at least in some case arose after apomictic seed formation.

Thus in several of the properties discussed the nine families resemble the families raised from the aberrants obtained. But that does not imply that the mother plants of these nine families were aberrants.

They may have originated from some later generation of the aberrants. The relatively good vitality characterizing some of these families, a characteristic in which they did not differ so much from the apomictic families, may be partly due to a selection among aberrants and their progenies. A plant of *Poa pratensis* possessing good vitality but poor seed setting (due to sexuality) is capable of surviving in nature for some time by vegetative propagation (shoots from rhizomes). On the average, however, the sexual families examined can hardly be considered capable of competing successfully against the apomictic families and they have not the same possibility of distribution in nature, partly owing to their lower seed setting, partly owing to the occurring segregation of less vital types. Thus the sexual plants in nature were assumed to be secondarily sexual, i. e. arisen from aberrants originated from apomictic strains. It seems hardly probable, although it cannot be proved, that any of them was derived from an original sexual population.

D. MORPHOLOGY, CHROMOSOME NUMBER, ETC. OF THE FAMILIES WITH A HIGH DEGREE OF APOMICTIC SEED FORMATION.

The 79 families having a high degree of apomictic seed formation exhibited a very great *morphological* variation between the families. There were hardly two families that were exactly alike. In many cases several families originate from one and the same locality. Thus, for instance, 8 types were collected at Offer in Ångermanland. Five of them were characterized as short at their habitat, the remaining three as tall. All the 8 families were morphologically quite different from each other, when compared under similar conditions of growth. In a meadow in Östergötland seeds were collected in two places having somewhat different environments. The plants obtained from each place showed a great variation among themselves, which also appeared in their chromosome numbers. Two plants from one of the spots had $2n = \pm 64$ and ± 77 respectively. There was also a difference between the two places. This could be shown, among other features, in the spreading capacity. There were 12 plants from each place and the average spreading capacity was the same, or 101 cm. But the variation between the plants from one of the places was considerably greater than between those from the other. σ^2 was 1117,0 and 72,0 respectively ($P < 0{,}001$). The same thing was true of a material collected in the meadows on the shore of Hallands Väderö. This extensive morphological variation agrees very well with the enormous formation of

variants that may occur, as shown above, owing to the various ways in which aberrants can arise.

It has, however, been possible to refer a number of families collected in different environments to certain morphological main types. This could be done, for instance, in the case of some families from the west coast of Skåne, which were referred to the form complex *irrigata*, and also certain families from central Germany, which have been placed in the form complex *angustifolia*.

Chromosome numbers. — Chromosome counts were made in 57 of the 79 families (Table 26). For all the investigated families the chromosome numbers are distributed fairly equally among the classes 50—60, 60—70, 70—80 and 80—90. The chromosome number of six of the families was above 90. The highest number found was $\pm$ 124, counted in a family collected in East Prussia. The lowest number observed in these families was $\pm$ 50. In Table 26 the chromosome numbers have also been separated according to origin. It is quite evident from the table that the chromosome classes are not equally represented for the regions recorded. In families from Germany, for instance, most of the numbers are below 60, while those from England have all above 80. That this is probably not due to chance will be discussed in the next chapter, where the chromosome number will be associated with the morphological type and habitat.

BROWN (1939) has made a list of the chromosome numbers of *Poa pratensis* known at that time. He found $2n = 56$ more frequent than other numbers. His list and results are, however, not quite correct, as his numbers can be assigned not only to investigated plants but to different values obtained from one and the same plant. Moreover, as pointed out by MÜNTZING in 1933, all old data of euploid numbers are probably not correct.

A new list is given in Table 28, and is based on the values for the *apomictic* strains in Tables 2 and 26 and on the material published by MÜNTZING (1933 a and 1940), RANCKEN (1934), NISSEN (1937), FLOVIK (1938), SKOVSTED (1939) — Scandinavian investigations —, ARMSTRONG (1937), BROWN (1939) — American investigations —, STÄHLIN (1929) and AVDULOV (1931). Thus the table does not include 1) strains known to be sexual, 2) aberrants, and 3) a few apomictic strains of our own material, the chromosome numbers of which have not yet been determined carefully enough. The material published by MÜNTZING (1940) and SKOVSTED (1939) consisted of twins obtained in seed testing. In the material examined by SKOVSTED in particular several twins had

TABLE 28. *Chromosome numbers found in Poa pratensis (aberrants and sexual plants not included. For origin of numbers see text).*

Chromosome number ± and number of plants															
2n	No. of plants	2n	No. of plants	2n	No. of plants	2n	No. of plants	2n	No. of plants	2n	No. of plants	2n	No. of plants	2n	No. of plants
39	—	46	—	53	1	60	2	67	5	74	2	81	1	88	1
40	—	47	—	54	6(5)	61	1	68	9(8)	75	2	82	3	89	2
41	1	48	11(2)	55	—	62	2	69	1	76	2	83	3	90	—
42	2	*49*	5(3)	*56*	9	*63*	—	*70*	5	77	4	*84*	2	*91*	1
43	—	50	20(14)	57	—	64	9(8)	71	—	78	2	85	2	92	—
44	—	51	2	58	1	65	2	72	5	79	1	86	4	93	—
45	3	52	4(2)	59	—	66	4(3)	73	1	80	1	87	6	94	2
Total	6	—	42(21)	—	17(16)	—	20(18)	—	26(25)	—	14	—	21	—	6

Moreover 1 plant with 2n = ± 28, 1 plant with 2n = ± 98, 1 with 2n = approximately 98—100 and 1 with 2n = ± 124.

the same somatic number (e. g. 9 with 2n = 48 and 7 with 2n = 50). It is probable that several of these values originate from one and the same morphologically uniform strain and should therefore be recorded only once. If that is done for all the twin plant materials of MÜNTZING and SKOVSTED we shall obtain the values given in brackets in the table. Altogether 156 plants have been investigated, but they probably represent at most 131 morphologically different types. For one form of *angustifolia* AVDULOV mentions the number 2n = 28, the lowest number so far known in *Poa pratensis*. Only a few values fall below 2n = 48, most of them from North America. All numbers between 49 and 91 are not represented, it is true, but they probably exist, as the values recorded are ± values. It is noteworthy that the euploid numbers (7 is the basic number in *Poa*) are not represented by more plants than the aneuploid, and the euploid numbers ± 1, ± 2 and ± 3 seem to be equally frequent. The number of plants with 2n = ± 50 is somewhat greater than those having other values. This is possibly only a chance occurrence. There are only 6 cases, all from our own material, in which the 2n was above 91, and the above-mentioned value of 2n ± 124 is the highest known so far from nature. Judging from the material recorded here, 49 seems to be the lowest euploid number, from which the aneuploid forms have as a rule originated.

Pollen formation and seed setting. — Only a small part of the material has been investigated with respect to pollen formation. In most of the examined plants the stamens were well developed. Only 2 out of 14 investigated families showed an evidently impaired stamen formation. Both these families were collected in Norway. In one of them practically all stamens were shrivelled. Seed setting in bagging was also nil, a few seeds were obtained in isolation by distance, and in free flowering with access to other pollen the seed yield was about 80 per cent. Other investigations, too, show that this type requires pollination to produce seed. It constitutes a parallel case to strain 701 described previously, but is possibly not so extreme.

Thus there exist in nature practically male sterile apomictic types of *Poa pratensis* that are pseudogamous. In this respect they are to a certain extent reminiscent of the form *caespitans*, which is entirely male sterile, reported in *Poa arctica* by NANNFELDT in 1940. In that case NANNFELDT, however, considers pseudogamy to be improbable, and that seems to be justified in view of the origin of the form, etc. But male sterility alone does not exclude the occurrence of pseudogamy.

Of 24 investigated families 16 had more than 90 per cent morph-

ologically good pollen, and for the other 8 the lower limit for the pollen quality was 70 per cent. Thus the quality of the pollen is throughout satisfactory. The low values for pollen quality most frequently occurred in families with low chromosome numbers, $2n < 60$, but the differences between the chromosome classes were not great. The material does not, however, contradict a previous observation (v. MÜNTZING, 1940) that the partially pollen sterile types are found among those with the lowest chromosome numbers.

Seed setting in isolation in the apomictic strains was also subject

TABLE 29. *Seed setting in isolation for all the material investigated in 1937.*

2n =	Percentage seed setting 0	20	40	60	80	100	Total number of plants
50—60	6	2	—	1			9
60—70	1	1	2	1			5
70—80	2	1	1	—			4
>80	6	2	8	1			17

to great variations. A number of factors influencing the seed setting in isolation has been discussed previously. Here we shall only touch upon the seed setting for different chromosome numbers. A satisfactory comparison of this kind can only be made with the material investigated in 1937. Table 29 gives a classification of *all* the material

TABLE 30. *Average seed setting in families with 2n = 50—60 and 80—90 in 1937.*

2n =	Number of families	Percentage seed setting Isolation	Free flowering
50—60	9	$21{,}53 \pm 6{,}63$	$67{,}87 \pm 3{,}98$
80—90	15	$32{,}20 \pm 5{,}09$	$78{,}65 \pm 2{,}04$

from that year, when at least two plants in each family were isolated. The table conveys the impression that the high chromosome numbers show a higher seed yield and an increased variability. If an examination is made in the classes $2n = 50—60$ and 80—90 (Table 30), neither higher seed setting nor increased variability can be shown for certain. A difference in seed setting exists but it is not significant. In view of the observation made that a low chromosome number is more frequently accompanied by an inferior pollen quality than a high chromosome number, a difference might have been expected. That such a difference could not be shown for certain may be due to the numerous

factors, already mentioned, which influence not only the *quality* of the pollen but also have an effect on the seed setting after isolation.

In Table 27 are presented data for the seed setting in *free flowering* for 50 apomictic strains investigated in 1937 and 1938. Seed setting in free flowering was not subject to so great a variation as that in isolation and the average for the said years was about 77 per cent. In the 1937 material a comparison has also been made between the seed setting in free flowering for $2n = 50—60$ and that for $2n = 80—90$. The difference is approximately equal to that in isolation and is demonstrable ($0{,}05 > P > 0{,}01$). Such a difference may be due to different causes, but it appears probable that here, too, we have an influence of the inferior pollen formation which on an average characterizes families with a low chromosome number. As already pointed out, these families depend for their seed setting on the supply of pollen from other plants.

TABLE 31. *The 1000-seed weight for all material investigated in 1937.*

	1000-seed weight in mg.					Total number
2n =	300	400	500	600	700	of plants
50–60	6	3	—	—		9
60—70	5	—	—	—		5
70—80	1	2	1	—		4
>80	2	7	7	1		17

Table 31 gives an idea of the variation in the *1000-seed weight*. The table is based on the results obtained in the 1937 material.

A considerably higher 1000-seed weight is shown by plants with the higher chromosome number. In the chromosome number class 50—60 the average 1000-seed weight was 382 mg., while in the class 80—90 it was 484 mg. (Diff. 102 mg. $P < 0{,}001$). A striking feature in this investigated material is that the 1000-seed weight showed an evident tendency to increase at first in $2n = 70$ and above.

Above we showed the behaviour of a number of properties in different chromosome values. It should finally be emphasized, however, that the origin of the material does not permit us at once to say that the changes *were due to the high chromosome number*. But that that is probably the case will be shown in the following chapter.

The investigations of the *aberrants* obtained from the apomictic families were not so extensive. Everything indicates that as regards origin, vitality and seed setting and the variability and absolute values

of these properties, they do not essentially differ from the aberrants obtained from the 12 strains described in the foregoing pages. The average height of 16 sure aberrants was $57{,}5 \pm 4{,}23$ cm., while for the corresponding maternal plants it was $70{,}7 \pm 1{,}36$ cm. The variance quotient for the inter- and intravariation will be $4{,}80$ ($0{,}05 > P > 0{,}01$). The seed setting in free flowering was examined in 12 such aberrants. It was found to be on the average $65{,}3 \pm 7{,}02$ per cent as against $84{,}3 \pm 2{,}42$ per cent for the maternal plants, variance quotient $= 7{,}12$ ($0{,}05 > P > 0{,}01$). The aberrants seem to have a lower height and an inferior seed setting and to show a greater variation in these properties than the maternal plants.

3. RESULTS FROM CROSSES WITH POA ALPINA.

A. INTRODUCTORY NOTES.

A number of types, assumed to be hybrids between *Poa pratensis* and other *Poa* species, have been described from nature. In Scandinavia such hybrids are known only from localities in the far north. Plants described as hybrids between *Poa pratensis* f. *alpigena* and *P. alpina* and also between f. *alpigena* and *P. arctica* R. Br. have been found very frequently. As *arctica* is not included in our material, the latter hybrids will not be discussed here. The mountain forms of *pratensis* × *alpina* found are all viviparous. Nannfeldt (1937), however, mentions a plant, found at Uppsala, which was probably a hybrid but was not viviparous. According to Smith (1920) and Nannfeldt (1937), the hybrid forms from the mountains are characterized by a certain, although not great, variation. The hybrid is occasionally recorded under the name of *Poa herjedalica* H. Sm.

As mentioned above, in the artificial crosses with *Poa pratensis* several different *Poa* species, but principally *P. alpina*, were used as father plants. Before discussing these crosses, a short summary will be given of the progeny obtained with other *Poa* species. The chromosome numbers found in these species will also be mentioned.

Pollination was performed with the pollen of *Poa annua* L., but as the cross was not made with emasculated flowers the positive effect of the pollination could not be studied. No hybrids were obtained.

Attempts were made to cross with *Poa trivialis* L. but were invariably unsuccessful owing to the impossibility of obtaining pollen of *trivialis* in the greenhouse. In five plants, two from Norway, the number $2n = 14$ was obtained, which therefore agrees with other reports that *trivialis* is a diploid. A sixth plant was

trisomic with $2n = 15$. This plant was very weak. So far attempts to obtain pollen from plants of *Poa Chaixii* VILL. cultivated in the greenhouse have failed.

In crosses with the three species *Poa nemoralis* L., *P. palustris* L. and *P. glauca* VAHL, belonging to the same subgenus *Stenopoa*, no seeds were obtained in pollination with the first two. Two plants were obtained from the *glauca* crosses, both made with the sexual strain 709 as mother plant. One of these plants was a haploid, the other, judging from the chromosome number, was a hybrid. The 2n in the hybrid was approximately 71, strain 709 has ± 90, while the 2n for the father plant was ± 50 (counted by KIELLANDER). When planted out in 1940 this hybrid plant was very weak and has not yet developed any panicles, hence nothing can be said of its morphology. In another *glauca* plant $2n = \pm 65$ was found and the 2n value of the two *nemoralis* plants employed was ± 28 and ± 42 respectively. The numbers given for *glauca* and *nemoralis* were known previously.

Five plants were obtained after crossing with *Poa compressa* L. The analysis of these plants was complicated owing to the fact that the mother plant, which was at first assumed to have apomictic seed formation, was found to be an aberrant and therefore probably sexual. It has not been possible, morphologically, to determine for certain the hybrid nature of the plants. As the chromosome number of the aberrant is not known, the plants could not be controlled cytologically. They are nevertheless very probably real hybrids but must be further investigated.

Using *compressa* as mother and *pratensis* as father BRITTINGHAM (1941) succeeded in obtaining a hybrid, arisen, judging from the chromosome numbers, by the fertilization of an unreduced female gamete. — The 2n values for three *compressa* strains were ± 35, 42 and 49 respectively. The 2n values found in other investigations are 42 and 56 (ARMSTRONG, 1937), hence $2n = 35$ and 49 are new numbers.

Of the species studied there remain *Poa alpina* L. and *Poa bulbosa* L., both of which belong to the same subgenus *Subbulbosae* (according to NANNFELDT, 1935). Taxonomically, this subgenus is considered to be closely related to *Stoloniferae*. All the *Poa bulbosa* types were completely viviparous without any trace of flower formation and were therefore useless for crossing. The numbers $2n = \pm 28$ and ± 45 were obtained in two types. ARMSTRONG (1937) mentions only $2n = 28$.

Five different strains of *Poa alpina* were used in the crosses with *pratensis*. One of these strains originates from St. Gothard, Switzerland, three were obtained from Germany, one of them from the Botanical Gardens in Berlin-Dahlem, and the fifth was collected on the mountain Åreskutan (Sweden). One of the strains from Germany (Bavaria) is morphologically very interesting. The lateral shoots successively form panicles during the whole of the summer, and towards the end of the vegetative period panicles in all stages of development are found on the plants, from panicle shoots up to mature seeds. The four strains from Germany and Switzerland are morphologically constant, whereas the Swedish strain has not been examined in this respect. Not much is known as yet concerning the chromosome

number in these five strains. Only the specially described Bavarian strain has been determined for certain. It has $2n = 31$. According to MÜNTZING, in *Poa alpina* certain chromosome numbers seem to be predominant. But MÜNTZING does not mention any apomict with $2n = 31$, thus we have here a representative of a new group. MÜNTZING has also an apomict from St. Gothard, which had $2n = 37$. On account of the said observation of the division of the apomicts into certain chromosome numbers, we seem to be fairly justified in assuming that our own plant from the same locality also has $2n = 37$. The chromosome numbers found in the hybrids with this *alpina* strain closely agree also with those expected at $2n = 37$ for the male parent. For the same reason the chromosome number of the Swedish strain may be assumed to be 38. No values at all can be given for two of the *alpina* strains.

In examining the development of the female gametophyte in a strain from Pajala, Sweden, MÜNTZING found diplospory, probably with a formation of the E.S. according to the *Antennaria* scheme or in the same manner as in the apomictic types of *Poa palustris* (KIELLANDER, 1935 and 1937). The formation of the E.S. was also studied in the Bavarian strain in our own material with $2n = 31$, which seems to take place normally, at least, in the same manner as in MÜNTZING's Pajala strain. The E.M.C. increases greatly in size, large vacuoles are formed, and the first division appears to be quite mitotic. (Fig. 56.) As this strain was constant in its progeny it should have mainly apomictic seed development. This is probably also true of the other three strains which had constant progenies.

Hybrids between apomictic *pratensis* and *alpina* strains are of great interest theoretically, as they are hybrids between species in which the formation of the female gametophyte takes place by apospory in one case and by diplospory in the other case. One of the hybrids described below has already been discussed (ÅKERBERG, 1936 b). It was then in the F_1 generation while at present the F_3 generation has been raised. MÜNTZING (1940) mentions the chromosome numbers of some hybrids produced after crossing with a sexual *pratensis*. Most of them had arisen from a reduced egg-nucleus, but one probably arose from an unreduced egg-nucleus.

The results presented below from the study of the species hybrid *Poa pratensis* × *alpina* are not yet so extensive. But as some of them are of interest for the following general discussion they have been included in the present work.

B. F_1 PLANTS OF POA PRATENSIS × ALPINA.

Origin, morphology, etc. — All hybrids were obtained from crosses in which *pratensis* was used as mother and *alpina* as father. It has already been mentioned that seed setting in *pratensis* after pollination with pollen from *alpina* was as good as that after pollination with *pratensis*. As far as the *apomictic* strains are concerned, the progeny after pollination with *alpina* was also mostly maternal, but in pollination of the *sexual* strain 709 hybrids always arose. 584 emasculated flowers of strain 709 were pollinated with pollen from *P. alpina*, from which 22 seeds were obtained, while 622 flowers pollinated with pollen from *P. pratensis* also gave 22 seeds, i. e. hybrids with *alpina* occurred as often as cross products in *pratensis*. Hybrids alone were also obtained in crossing with aberrants from *pratensis*, which according to what has been shown above are sexual. Thus, from an aberrant having $2n = \pm 66$, crossed with *alpina* with $2n = \pm 37$, a hybrid with $2n = \pm 54$ was obtained. The hybrids arising in crosses with apomictic strains were not less rare, comparatively, than the pure *pratensis* aberrants.

To draw any conclusions from these crossing experiments as to the occurrence of hybrids in nature the following points must also be borne in mind. *Poa alpina* flowers much earlier than *pratensis*, in the cultural experiments two—three weeks earlier. Hybridisation in nature should therefore be possible less frequently. In the artificial pollination experiments flowering *alpina* was taken from plants in the open field, while the *pratensis* plants were grown in a greenhouse. The frequency of hybrids between apomictic types of *pratensis* and *alpina* can never be very high, and moreover, as will be shown below, some hybrid plants have certainly no possibility of prevailing in nature owing to their low vitality. NANNFELDT (1937, p. 20) states, however, that the hybrid is »relatively common within large areas».

Altogether 19 hybrids were obtained and they originate from 9 different *pratensis* plants and, as mentioned above, from 5 different *alpina* plants. The chromosome numbers were determined in 16 of these hybrids, and judging from the numbers obtained 12 arose from reduced egg-cells and 4 from unreduced. Thus the hybrids arise in a similar manner to the pure *pratensis* aberrants, i. e. as »diploid» or »triploid» hybrids. One of the »triploid» hybrids had strain 709 as mother plant, and this is the previously discussed case with an *unreduced* fertilized egg-cell from that strain (♀ $2n = \pm 90$, ♂ $2n = \pm 37$; F_1 $2n = \pm 108$). Four other hybrids from strain 709 gave $2n = \pm 70$,

± 66, ± 70 and ± 66 respectively (the 2n for the ♂ was here ± 38). Thus these four hybrids arose by the fertilization of a reduced egg-cell. The chromosome number of the 16 hybrids varied between ± 41 and ± 108.

On account of their mode of origin the artificially produced hybrids should be expected to show a great morphological variation. As extreme cases we may compare two of the hybrids obtained, one »diploid» with 2n = ± 41 (♀ 2n = ± 50, ♂ 2n = ± 31) and the other »triploid» with 2n = ± 108 (♀ 2n = ± 90, ♂ 2n = ± 37). In the former case the ratio of *pratensis* and *alpina* chromosomes is approximately 1 : 0,62, in the latter case the ratio is 1 : 1,21. *Pratensis* should be much more dominant in the characters of the latter plant than in the former. The

Figs. 54 and 55. Two *Poa pratensis* × *alpina* hybrids. To the left in both figures the mother plant, to the right the hybrid. Note the difference in vigour.

19 hybrid plants obtained also differed very much from each other and as many different types were found. In certain characters, however, the father plant (*alpina*) was very pronounced, almost independently of the number of *pratensis* chromosomes present. Quite naturally, however, the dominance was frequently not so marked in the high chromosomal F_1 plants. As an instance of such a dominance from *alpina* we may mention the tuft formation. Thus all F_1 plants developed tufts intensely without rhizomes, even if the tufts were not so compact as in *alpina*. In seven F_1 plants the average circumference of the tufts in the second vegetative year was 37 cm., while in the corresponding *pratensis* (female) strains it was 115 cm. As a rule the F_1 plants were clearly earlier and shorter than *pratensis*, but this difference was not discernible in a few high chromosomal plants. Another typical feature was that the culm leaves were longer and the spikelets larger and of a different shape in the F_1 plants than in *pratensis*.

None of the F_1 plants showed any tendency to vivipary, nor was vivipary observed in any progeny material of these plants. Some of the plants were very weak, in fact so weak that they could not have survived in nature. Others again were vegetatively as vigorous as the parents (Figs. 54—55). The very weak plants were all »diploid» hybrids.

Formation of male and female gametophytes. — No cytological investigations of the division in the P.M.C. have been carried out as yet, but examinations have been made of the quantity and quality of the pollen. As a rule the stamens were well developed. The pollen of certain F_1 plants was morphologically as good as that of the *pratensis* (♀) strains, in other cases it was inferior. Both types occurred among the »triploid» and among the »diploid» hybrids. The following scheme shows the distribution of the investigated plants in different classes.

	Percentage morphologically good pollen						
	0	10	30	50	70	90	100
F_1 plants	1	2		3	4	1	

The pollen of the F_1 plants was strikingly large, larger than that of the *pratensis* strains and even larger than that of *alpina* in those cases it was examined. MÜNTZING (1940) gives the pollen size for both *Poa pratensis* and *P. alpina*, and according to his measurements the pollen of *alpina* seems not to be larger on an average than that of *pratensis*. The size of the pollen in the hybrids therefore appears to differ from that of both parents, and that is true of the »triploids» as well as of the »diploids».

This increase in the size of the pollen seems to be at least partly a sequel of the formation of hybrids, i. e. due to the gene combinations arising through the hybrid formation. Even if the chromosome number itself, as generally happens, involves differences in pollen size in comparison between different hybrid plants, it cannot be associated with the increase met with throughout. All the »diploid» hybrid plants, for example, have lower chromosome numbers than the *pratensis* (♀) strains. Such high values for pollen size as the highest shown by the F_1 plants were not, generally speaking, found in *pratensis* and *alpina*. The increase in pollen size, however, is also probably associated with the course of the reduction division, for the pollen of the F_1 plants is often very irregular in size. The following examples, in which the chromosome numbers of the female, male and F_1 plants are known, may illustrate this.

Of the F_1 plants recorded in the table below the »diploid» is the least variable with respect to pollen size, but that was not always the case. There were »diploid» plants that showed a very great variability. In some plants, on the other hand, the variability was not greater than in the parent strains. It may also be pointed out that the variation in pollen size found in these hybrids was not met with in the pure *pratensis*

TABLE 32. *Pollen size in Poa pratensis and P. alpina and in F_1.*

		Pollen size in μ				
P. pratensis	*P. alpina*	F_1 plants				
		»diploid»			»triploid»	
		1	2	3	1	2
30,8	35,5	36,9			39,6	50,9
31,7		39,4				
31,1		34,2	34,7	36,5		
35,0		47,5				
33,7		40,8				

aberrants. — This investigation of the pollen size points to a great irregularity in the division of the P.M.C., probably with the formation of unreduced pollen grains in certain hybrid plants, but it may also imply the probable existence of a considerable variation between different plants with respect to the course of the reduction division.

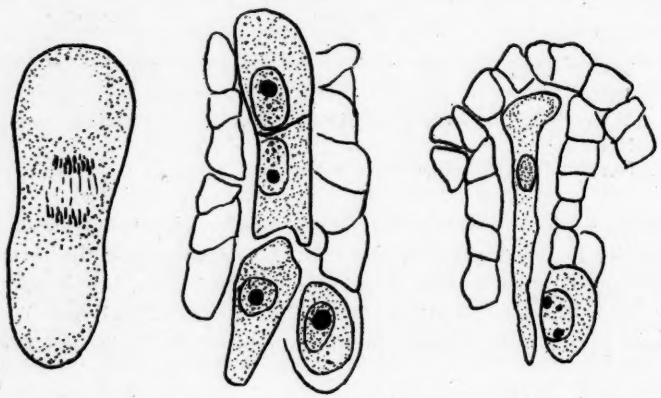
TABLE 33. *Pollen size in three F_1 plants compared with the parent plants.*

	2n	Pollen diameter, size classes												Pollen size		
	±	5	6	7	8	9	10	11	12	13	14	15	16	n	μ	σ^2
701 ♀	50	61	135	35	2									232	30,81	11,424
G 135 ♂	37	2	64	119	29	1								215	35,48	12,461
F_1 : 1	45	4	42	107	54	9								216	36,88	18,404
F_1 : 2	65—70	6	32	75	65	34	13	—	2	1				228	39,64	43,692
F_1 : 3	69	—	—	4	24	21	17	14	1	3	1	2	4	91	50,91	125,888

The formation of the female gametophyte was controlled to a certain extent in two of the F_1 plants. One of these plants was a »diploid» hybrid ($2n = \pm 41$; ♀ $2n = \pm 50$; ♂ $2n = 31$). The chromosome number of the other is unknown. The embryological material is not yet extensive enough to furnish a perfectly reliable picture of the development of the embryo-sac. The E.M.C., however, seems to undergo reduction division, very disturbed as far as can be judged.

Megaspore formation has been established for certain, and the occurrence of more than two megaspores indicates that reduction division has taken place (cf. KIELLANDER, 1937). In one case the chromosome number could be satisfactorily counted at division in an antipodal nucleus; it was approximately half the somatic number. In a flower (Fig. 58) a highly degenerated E.M.C. or possibly highly degenerated megaspores were observed. It was not possible to ascertain whether megaspores had had time to develop here.

Apomictic plants of *Poa pratensis* and *P. alpina* were used as



56 57 58

Fig. 56. Anaphase in the E.M.C. of a *Poa alpina* strain. — Figs. 57—58. From a hybrid *P. pratensis* × *alpina*. — 57. The dyad stage and two aposporous E.S. initials. — 58. A degenerating E.M.C. (see text) and an aposporous E.S. initial.

female and male parents of the F_1 plants embryologically investigated. As the F_1 plants appear to have reduction division in the E.M.C. and a possibility of forming aposporous embryo-sacs, the female gametophyte formation ascertained in *pratensis* is consequently the predominant one in these hybrid plants, and this also in »diploid» hybrids. The origin of diploid legitimate embryo-sacs, in these instances diplospory, seems to be recessive to the formation of embryo-sacs with reduced chromosome numbers. The result agrees with MÜNTZING's (1940) crosses in *Poa alpina* inasmuch as he found the F_1 sexual in crossing apomictic × sexual strains. Further information of the sporophyte formation in the hybrid plants is furnished by the F_2 (see below).

Seed setting. — Like other properties, seed setting in the F_1 plants

was variable. In most cases, however, it was low in isolation and in free flowering (cf. Table 34).

TABLE 34. *Seed setting in isolation and free flowering in 14 F_1 plants from the cross Poa pratensis × alpina.*

	Percentage seed setting							
	0	10	30	50	70	90	100	n
Isolation	11	2	1					14
Free flowering	6	3	2	2	1			14

Plants with poor or with very good seed setting were found among »diploid» and »triploid» hybrids, but the lowest yield was most frequently obtained among the »diploid» hybrids. A markedly good seed yield was, however, shown by one or two plants, a yield that was not inferior to that of the corresponding *pratensis* (♀) strain. Thus, as far as seed setting is concerned, these F_1 plants behaved in the same manner as the pure *pratensis* aberrants, even if the average yield was lower than that of the aberrants. — Plants with poor pollen had throughout a poor seed yield both after isolation and after free flowering, but that is also true of an occasional plant with morphologically good pollen.

C. F_2 AND BACK-CROSS FAMILIES.

Some of the hybrids obtained were tested in F_2. The origin of these hybrids is shown in the following table.

TABLE 35. *Origin of hybrids between Poa pratensis and P. alpina, tested in F_2.*

♀		♂		F_1 plants		2n = ±	
pratensis strain	2n = ±	*alpina* strain	2n = ±	1	2	3	4
701	50	G 135	37	65—70 [1]	45	?	69
702	50	G 44	31	41			
5303	78	G 43	?	50—55 [1]			
746	80	G 121	?	95	102		

Thus »diploid» as well as »triploid» hybrids were tested. The chromosome numbers of three of the F_1 plants were not counted, but the number of two of them can be given approximately according to the values obtained in the F_2.

The seeds from a few F_1 plants have been examined rather thoroughly as to germinability. It was found (ÅKERBERG, 1938) that

[1] See text.

the seeds of these F_1 plants germinated much quicker than seeds of *pratensis*. The rate of germination in *alpina* is much higher than in *pratensis*, and the F_1 plants were evidently intermediate in this respect. The total germination of these F_1 plants was approximately as high as in *pratensis*. The frequency of polyembryony was strikingly low. It seems to be higher, however, in the two F_1 plants from strain 746 than in the others. As regards the chromosome number present, *pratensis* is more dominant in these two plants than in most of the others described.

The F_2 of two F_1 plants and back-cross families will be described a little more in detail. One of these F_1 plants is the »diploid» hybrid having $2n = \pm 41$ from strain $702 \times$ G 44 which has been subjected to embryological investigation. G 44 is the apomictic *alpina* strain with continuous panicle formation described above. Seeds of the F_1 plant, obtained after isolation, free flowering and back-crossing with *pratensis* and *alpina* (G 44), were sown. The progeny thus obtained was very variable, which was ascertained morphologically and in chromosome determinations. In plants raised after isolation the following chromosome numbers were obtained: $\pm$ 1) 50 (see below), 2) 41, 3) 35; after free flowering, $\pm$ 68; after back-crossing with *pratensis*, $\pm$ 73, and after back-crossing with *alpina* (G 44), $\pm$ 1) 38, 2) 36, 3) 47, 4) 39, 5) 35, 6) 40. In most cases reduced female gametes seem to have functioned. The plant obtained after free flowering, and possibly also the one after back-crossing with *pratensis*, very probably arose from unreduced egg-cells. These results seem to agree very well with the embryological findings, the latter indicating the occurrence of embryo-sacs with reduced or unreduced chromosome number. The latter were then assumed to have an aposporous origin, but it may also be possible that they arose by diplospory. *Thus the F_1 plant just described, which was obtained from facultatively apomictic pratensis and alpina, has mainly sexual seed formation.*

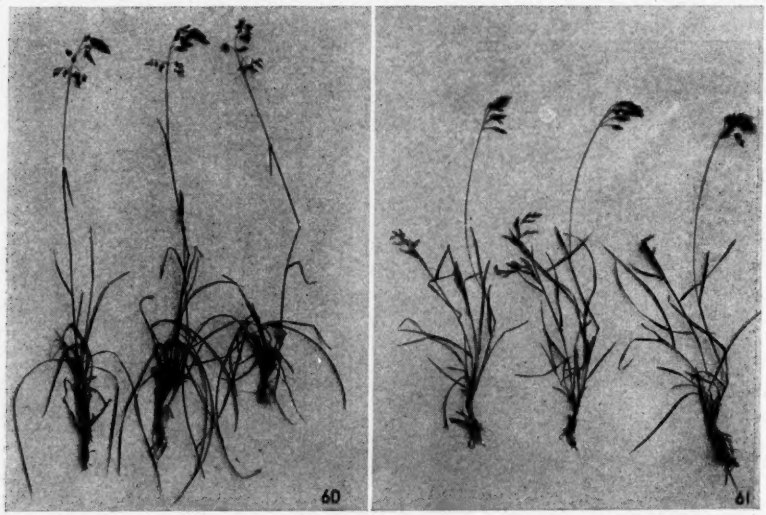
The morphological variation in F_2 and back-cross families was great. Some plants were weak in vitality and fertility, but in the back-cross with *alpina*, which was much more successful than with *pratensis*, the majority of the plants had good vitality and very good fertility. All plants, except one, were tuft-producing without any real capacity to form rhizomes. The exception was the plant having $2n = \pm 50$. This plant was identical with the mother strain 702 morphologically and in chromosome number. It might be assumed that an admixture had taken place, but the material was controlled in such a manner that this assumption does not seem justified. In order to explain the mode of origin of the plants, however, further material is required. The back-cross family with *alpina*, which was obtained by

pollinating the F_1 plant without previous emasculation, consisted of some thirty plants. Most of the plants could be referred to a certain main type, agreeing fairly well with the F_1, but there was a variation between the plants. The seed setting was about 30 per cent. Other plants could be divided into three types differing from each other and from the main type. One of these type-groups contained four plants, all of which possessed the peculiar property of the father plant, i. e. the lateral shoots continuously form panicles. Three of the plants have been controlled in the following generation. The families thus obtained were constant and alike and had the same chromosome number as G 44. It is certain that no admixture has taken place here, for no seeds of G 44 were sown at the same time as the back-cross family. If these three plants have arisen by fertilization a very curious gamete

Fig. 59. Three plants from a hybrid *Poa pratensis* × *alpina* back-crossed with *alpina*. To the left a plant, in which the lateral shoots continuously form panicles.

formation and heterogamy are present. Male parthenogenesis has been supposed to occur in certain cases with a complicated chromosome constitution (v. DARLINGTON, 1937, p. 193), and all the controlled properties in the three plants described point to such a mode of origin. In that event, however, *unreduced* pollen grains have functioned. But there remains the fourth plant with $2n = 47$ instead of $2n = 31$ as in the others. This fourth plant differed also quite clearly in certain morphological characters. That it must have arisen by the union of two gametes seems to be necessary, for instance, by a diploid male gamete and a female gamete with 16 chromosomes. Of the two remaining types, one was rather closely allied to the main type, but the other differed widely in vitality, fertility, colour, etc. Neither of them could be distinguished by chromosome number, but they are probably the result of a certain chromosome combination. It thus appears quite plainly from the above material that in the F_1 plant described we have a very complicated inheritance.

The 2n of the other F_1 plant that we shall specially describe was $= \pm 69$ (φ $2n = \pm 50$, σ $2n = \pm 37$). It was therefore a triploid hybrid, and families obtained from this plant after free flowering and back-crossing were planted out. These families, too, were clearly variable, which was established morphologically and by chromosome counts. After free flowering the following 2n values were obtained: 1) ± 65, 2) ± 65, 3) ± 70, 4) ± 72, 5) approximately 90, 6) ± 78. The plant with 2n approximately 90 probably arose by the fertilization of



Figs. 60—61. Two of the plants in Fig. 59. To the right the plant with panicles on the lateral shoots.

an unreduced egg-cell. The F_1 plant under discussion did not show so peculiar inheritance phenomena as the preceding one. A segregation in a certain character could be ascertained by selection at the seedling stage. Early and late seedlings were divided into three classes, and the following measurements (Scheme 7) of plant height were obtained in these classes. I denotes the first seedlings appearing.

The plants in class III are evidently shorter than those in class I. This difference in height is probably due mainly to a difference in vitality.

All the other F_1 plants, except one, controlled in F_2 showed a distinct morphological variation. The chromosome numbers of a

Scheme 7. Segregation in plant height in F_2.

Earliness class	Plant 1	2	Height in cm. 3	4	5
I	43	42	25	34	—
II	30	39	died	(10)[1]	—
III	15	died	18	19	(10)[1]

number of plants of these F_2 families were investigated, the following values being obtained (Scheme 8).

Scheme 8. Chromosome numbers in F_2.

F_1 $2n \pm$	Plant 1	F_2 $2n = \pm$ 2	3	4	5
?	approx. 60	56	65	68	65
47	» 50	65	75		
?	45	54	52	52	54

Two progeny plants of the F_1 plant with $2n = \pm 47$ have rather high chromosome numbers, but as both of them were obtained after free flowering it is rather difficult to control their origin. They probably arose by the fertilization of an unreduced egg-cell. Many plants in these F_2 families have a poor vitality. Solitary very vigorous plants, however, were met with, and also a few plants having short rhizomes. Like the seed setting in free flowering, the pollen fertility of the investigated plants varied very much, but several plants had a markedly high pollen fertility and good seed setting.

The family not showing an evident morphological segregation was derived from an F_1 plant with $2n = \pm 102$ (♀ $2n = \pm 80$, ♂ unknown). A few plants deviated, but the majority agreed morphologically. It remains to be seen whether we have here a case of highly apomictic seed formation in an F_1 plant or only a slight segregation. In a sister plant of this F_1 with $2n = \pm 95$, the segregation was very conspicuous.

D. SOME CONCLUSIONS.

The above described hybrids between *Poa pratensis* and *alpina* differ from species hybrids in general in that every hybrid plant produced differed in its chromosome number from other plants of the same kind, as a result of the chromosome numbers of the parents, their origin, etc. Therefore nothing general can be said of the hybrids, as may be often said of a hybrid between two species with a constant chromosome number. Instead each hybrid plant may be expected to

[1] No panicles.

behave in its particular manner with respect to characters, inheritance, etc. A similar hybrid variation has also been obtained in crosses between different chromosomal races of *Rubus* (CRANE and THOMAS, 1940 b). But it should also be borne in mind that in *Poa pratensis* and *alpina* the mechanism of embryo formation is not the same, which must further complicate the inheritance of the hybrid. It is therefore rather surprising that the seed setting even in an occasional »diploid» hybrid was relatively good. Things must be different in the »triploid» hybrids, in which all chromosomes of the *pratensis* plant are present.

A characteristic feature of all the artificial hybrid plants was the formation of tufts, in which property they are easily distinguishable from *pratensis*. This is also in agreement with observations of hybrid plants in nature (SMITH, 1920, and others). With but one exception, the latter were, however, viviparous, whereas vivipary was not observed in any artificially produced hybrid. The parent plants used in the artificial crosses were also all non-viviparous, while viviparous *alpina* plants are known from the alpine region where viviparous hybrids were met with. It is therefore probable that in the latter we have a gene conditioned vivipary, as also assumed by NANNFELDT (1937). Crosses between forms of *pratensis* and *alpina* growing in the vicinity of a viviparous hybrid might furnish proof of this assumption. If a non-viviparous hybrid arises in nature, however, and becomes sexual, in the same manner as artificially produced hybrids, it would be liable to back-crosses with its parents if its pollen production was low. Moreover, it segregates a large number of less vital types which cannot survive in nature, and will probably disappear gradually. That does not occur in the case of a vital viviparous hybrid, which is capable of holding its own and spreading. This also explains why the viviparous types are in the majority at the same time as they are relatively common.

Thus the primary hybrids between *pratensis* and *alpina* had mainly sexual seed formation, even when both parents were highly apomictic. As already pointed out, this is the result of a cross between species with respectively aposporous and diplosporous gametophyte formation. A number of functioning unreduced egg-cells have been observed, but it has not been possible to determine their origin. The possibility of the development of an embryo-sac with an aposporous origin has been proved embryologically. The results of the seed production shown in the hybrids also seem to be of great value in a discussion of the occurrence of apomixis in general and in *Poa pratensis* in particular.

III. DISCUSSION.

The development of the female and male gametophytes in Poa pratensis. — In the description of apomixis in *Poa pratensis* given in the preceding pages GUSTAFSSON's (1939 a) terminology was employed. The type of female gametophyte development found in apomictic plants of the species agrees entirely with apospory according to GENTCHEFF and GUSTAFSSON's definition (1940 b): the formation of a gametophyte from vegetative cells of a sporophyte by mitotic divisions. As *Poa* has only one archesporial cell, as is normal in the Gramineae, the derivation of the gametophyte from *vegetative* cells is easy.

As far as the apomictic phenomena in general are concerned, a rich terminology has been invented. An account of the origin and development of this terminology has recently been given, in addition to GUSTAFSSON (1935 and 1939 a), by STEBBINS and JENKINS (1939) and by FAGERLIND (1940). In the proposals for a terminology put forward by these authors, all of which are characterized by a careful differentiation of the gametophyte and the sporophyte development, there are certain differences with regard to the basis of classification, definitions and terms. But what was described above as apospory is also delimited by STEBBINS and JENKINS and by FAGERLIND, to which they apply the term somatic apospory. CRANE and THOMAS (1940 a) also classify different types of reproduction mechanism, but, as so often happened in earlier works, have again an intermingling of terms for the gametophyte and the sporophyte generation.

So far the only species of *Poa* in which apospory has been found is *pratensis*. In *Poa nemoralis, palustris, glauca* (KIELLANDER, 1935, 1937, and unpubl.) and probably in *P. alpina* (MÜNTZING, 1940, and present paper), on the other hand, diplospory and gametophyte development from a legitimate E.M.C. mainly take place, the E.M.C. dividing by so-called somatic division (GUSTAFSSON, 1939 a, and earlier). On account of the mitotic character of the division, this type of diplospory is called generative apospory by FAGERLIND. Thus, for FAGERLIND the basis of classification is the character of the division, for GUSTAFSSON, STEBBINS and JENKINS it is the origin of the gametophyte (somatic cell or archesporial cell). As far as *Poa* is concerned, the latter basis of classification seems to be more justified, among other things, because in this genus we have a very pronounced morphological difference between the archesporial cell (= E.M.C.) and vegetative cells.

The oldest example of apospory in species with one archesporial

cell was described in *Hieracium* subg. *Pilosella* (ROSENBERG, 1907). To that type, to which *Poa pratensis* most closely belongs, and which has been called »the *Hieracium Pilosella* type of stabilized apospory» by GUSTAFSSON (1939 a), we may also refer *Crepis* (STEBBINS and JENKINS, 1939), *Hypericum perforatum* (NOACK, 1939) and possibly also a few species of *Hieracium* subg. *Archieracium* (GENTCHEFF, 1937). A comparison between the development of the E.M.C. and that of the aposporous E.S. initials in these species is of special interest.

On the one hand mainly sexual types, on the other hand mainly aposporous types and also transitional types between these two were obtained in *Poa pratensis*. A similar and very distinct grading has also been obtained in different species and forms of *Hieracium* subg. *Pilosella* and *Crepis*. In the sexual forms and species there is a normal megaspore development, and a normally built embryo-sac arises from one of the megaspores. In *Poa pratensis*, however, only three megaspores are usually formed, the micropylar dyad cell degenerating before division. In the apomicts the development of the E.M.C. is very varied. In one species of *Crepis*, subsp. *pumila* apom. *hamiltonensis*, the E.M.C. never reaches beyond early prophase before it degenerates. In other forms of *Crepis* a degeneration has been observed in prophase and even later, and that also seems to occur in *Poa pratensis*. But it is not always that the E.M.C. or its products degenerate in the apomictic forms. Progeny investigations have shown that legitimate embryo-sacs may occasionally form embryos (haploid and diploid aberrants). A very extreme degree of degeneration was found in one of the embryologically investigated F_2 plants, in which not only the E.M.C. was disintegrated but probably also the entire nucellus, consequently no seeds were formed despite the occurrence of aposporous E.S. initials. In an *Archieracium* species GENTCHEFF (1937) observed degeneration when the embryo-sac was in a 1—8-nucleate stage. It seems probable that these forms of degeneration are a kind of diplontic sterility, an unequally effective physiological disturbance in different species and forms, causing the degeneration of the E.M.C. or its products. A similar process most probably occurs also in *Hypericum perforatum*, where the degeneration may affect the E.M.C. already in the prophase stage. NOACK, however, is of opinion that in this species the degeneration is partly a result of disturbances in the course of meiosis. In aposporous species with many-celled archesporia (the *Alchemilla* type of stabilized apospory; GUSTAFSSON, 1939 a) a degeneration of the axillar E.M.C., as well as that of secondary E.M.C:s, also takes place in early

prophase. This was the case, for instance, in species of *Alchemilla*, *Oxyria*, *Atraphaxis* (see GUSTAFSSON's list, 1939 a), an occasional *Rubus* species (CRANE and THOMAS, 1940 a), and others.

In *Hieracium* subg. *Pilosella* ROSENBERG (1907) describes a conspicuous competition between aposporous and legitimate embryo-sacs, which most frequently leads to an encroachment upon the legitimate embryo-sacs. Such a competition has also been observed in *Poa pratensis* and *Crepis*. When the aposporous E.S. initial emerges into the chalazal region, it acquires a greater ability to hold its own in the competition by having a more ready supply of nutritive material. The competition will also depend of course on the time when the aposporous E.S. initials appear in relation to the development of the E.M.C. In comparing the division in the E.M.C. and that in the P.M.C. in the apomictic strain 746, described above, and in the sexual strain 709, it was found that the development of the E.M.C. was later in strain 746. It is possible that this delay in the E.M.C. affords an increased competitive ability to the aposporous E.S. initials. On the other hand, the E.S. initials seemed to be delayed in the aberrants from the apomictic strains, and these aberrants were highly sexual.

In *Poa pratensis* the E.S. initials appear in the cell layer nearest the E.M.C. As yet we know nothing of how they arise or whether the E.M.C. has any influence on their origin. Their appearance, however, is certainly not caused by the degeneration of the E.M.C., for the E.M.C. does not always degenerate, and moreover there are plants in which the E.M.C. forms an embryo-sac in spite of the presence of aposporous E.S. initials. The F_1 aberrants with a high frequency of twins in all probability belong to the latter category, provided this formation of twins is the expression of the occurrence of aposporous E.S. initials. This is indicated by the embryological investigations. In that event we have here a certain analogy to »the *Leontodon* type of unstabilized apospory». In *Leontodon hispidus* and *Picris hieracioides* BERGMAN (1935 a) found plants with a tendency to the development of aposporous E.S., but they were unable to form embryos. The same thing characterizes *Coreopsis bicolor* (GELIN, 1934) and other species. The apospory found in *Leontodon hispidus* is hereditarily conditioned. Triploid plants have often been shown in twins of sexual species (see WEBBER, 1940). At present we know nothing for certain of the origin of these plants, but it seems probable, as MÜNTZING (1937 a) has already pointed out, that they, too, arise by the fertilization of incidental aposporous embryo-sacs. Since the frequency of triploids is not the

same in different species, some species may possess a hereditary tendency to form such embryo-sacs. The aberrants in *Poa pratensis* do not seem to be quite so extreme as the *Leontodon* case, for in the former the aposporous E.S:s are evidently able to form embryos.

The aposporous E.S. initials in *Poa pratensis* have a long period of growth before division. They differ very much in shape and appearance before the first nuclear division from a megaspore that will develop into an embryo-sac. This is probably often due to a change in location before the division. As already pointed out by GUSTAFSSON (1939 a), they agree in growth, etc. with *Hieracium pilosella* and *Crepis* and, as far as can be judged, also with *Hypericum perforatum*. The embryo-sac formed seems to agree entirely with respect to construction with a legitimate embryo-sac from an E.M.C., the only difference being that the former has the diploid, the latter the haploid chromosome number.

The occurrence of an aposporous or a legitimate E.S. in *Poa pratensis* thus seems to be connected with 1) a factor, or factors, which produce aposporous E.S. initials, 2) the time of the appearance of the latter, 3) a factor, or factors, which cause a degeneration of the E.M.C., and 4) possibly the course of the reduction division in the E.M.C. (haplontic sterility).

No degeneration of the P.M.C. in the same manner as occasionally seen in the E.M.C. in certain strains of *Poa pratensis* has been observed. The few data from the course of the reduction division in the E.M.C. and the P.M.C. from apomictic and sexual strains did not show any difference. Univalents were observed in all cases. With regard to the quality of the pollen in apomictic strains MÜNTZING (1933 a) points out that it is remarkably good. This observation is confirmed by the present investigations. Some strains, it is true, had a somewhat inferior quality, which was in addition easily changed modificatorily. Normally, however, it was seldom below 70 per cent, and it should be pointed out that such a poor quality as that found in several plants arisen after sexual seed formation was never met with. In some cases, however, the quantitative pollen formation may have been greatly reduced, and this in combination with poor quality (probably a combination of diplontic and haplontic sterility) resulted in a very low seed setting in isolation (e. g. in strain 701). It is not possible to give any average values for the pollen production in plants produced sexually and apomictically. The conspicuous superior pollen formation characterizing the latter is probably a result of selection, partly owing to the

fact that poor pollen quality was frequently associated with low vitality, partly because the apomictic plants also require pollen for seed setting (pseudogamy).

Other species of *Poa* with apomictic seed formation are also characterized by satisfactory pollen formation on the average. That was the case in *Poa alpina* (MÜNTZING, 1933 a, 1940), *P. palustris*, *P. nemoralis* and *P. glauca* (KIELLANDER, unpubl.). Pseudogamy has not been shown in these species, but it probably occurs in some strains of *alpina*. The good pollen production found in the apomictic species of *Poa* is contrary to the state of things in apomicts in general (see ROSENBERG, 1930), and the pollen production also seems to be somewhat better than in other pseudogamous species, e. g. *Potentilla* (MÜNTZING, 1928), some diploid *argentea* strain of which, however, was quite fertile, and *Hypericum perforatum* (NOACK, 1939). But such a degeneration of the division of the P.M.C. as that met with in certain total apomicts (*Taraxacum*, *Hieracium*) does not occur in either of these pseudogamous species.

The embryo and the seed. — In *Poa pratensis* it has been shown that the egg-nucleus in both the legitimate and in the aposporous E.S. can develop parthenogenetically and after fertilization, but that pollen is necessary for seed formation in *both* cases. In the former fertilization is the normal procedure, in the latter parthenogenesis. In *Hypericum perforatum*, even this species pseudogamous (NOACK, 1939), the egg-nucleus of the aposporous embryo-sac seems to be fertilized relatively often, especially in pollination with other species. BERGMAN (1935 a), too, found the occurrence of fertilized aposporous E.S:s in *Leontodon*. Sexuality has also been shown in aposporous gametophytes in a fern (*Scolopendrium vulgare*; ANDERSSON-KOTTÖ and GAIRDNER, 1936). Thus apospory is not inevitably accompanied by parthenogenesis in angiosperms and ferns, as seemed to be the case from earlier investigations.

A summary of the causes that may give rise to a parthenogenetic development in known cases of apospory has been given by GENTCHEFF and GUSTAFSSON (1940 a). Much was then very obscure with regard to the apomictic strains of *Poa pratensis*, but on the basis of the investigations reported in the present work, the seed formation at least in some of these strains may be characterized as autonomous parthenogenesis with a stimulation or fertilization of the central nucleus. Fertilization seems to be the more probable. As a rule the egg-nucleus of the aposporous embryo-sac has passed through several divisions before

the pollen nucleus reaches it. The material investigated so far indicated that if an egg-nucleus happens to be delayed in its development, as, for instance, in polyembryony, it is more frequently fertilized. Thus parthenogenesis is maintained by the ability of the egg-nucleus to begin to divide early. This development seems to agree most closely with that found by GENTCHEFF and GUSTAFSSON in certain strains of *Potentilla collina* (C—B and others).

The egg-nucleus in the aposporous embryo-sac in *Poa pratensis* may therefore be fertilized, but from causes, the genetic nature or origin of which cannot yet be explained, an embryo is usually formed before fertilization can take place. We thus find here only a change towards conditions in a sexual species, in which fertilization is the rule and parthenogenesis the exception. The origin of the latter is elucidated in a number of experiments to induce parthenogenetic development (haplo-parthenogenesis) in certain sexual species, when the development of the embryo in many respects is reminiscent of diplo-parthenogenesis in *Poa pratensis* and certain other apomicts. KIHARA (1940) pollinated a strain of *Triticum monococcum*, which normally gives 0,5 per cent haploids, with X-rayed pollen and obtained more than 13 per cent haploids. In late pollination with pollen from another species 6—9 days after emasculation 17,5 per cent haploids were obtained. Thus in these instances the central nucleus and not the egg-nucleus was fertilized. Endosperm formation was necessary for seed production. A summarised report of other similar cases of the occurrence of haploids was made by IVANOV in 1938. Several of these haploids arose by species crossings and it is conceivable that their appearance is associated with a slower rate of germination in species-foreign pollen. — That the occurrence of haplo-parthenogenesis in sexual species is very rare, however, may also be due to other properties in the haploid embryo, which have nothing to do with the ability of the egg-nucleus to develop parthenogenetically (cf. DARLINGTON, 1939, p. 108).

If the central nucleus in apomictic strains is fertilized, the chromosome ratio in the embryo, endosperm and soma differs very much from that in sexual species, nor is it constant (for instance, we may pollinate a strain with $2n = 50$ with a father plant having $2n = 50$ or 90). But as already pointed out by GENTCHEFF and GUSTAFSSON (1940 a), this relation does not seem to have such a great influence at least on certain apomicts as on sexual species (MÜNTZING, 1930 b, 1933 b). It is also possible that the occurrence of parthenogenesis has a certain connexion with this phenomenon.

No fertilization of the aposporous egg-nucleus has been shown in the apomictic species of *Crepis* and *Hieracium pilosella*. These species, however, are not pseudogamous, but possess a completely autonomous apomixis with an early embryo formation. But in certain facultatively apomictic *Hieracium* species the legitimate (haploid) embryo-sacs may be fertilized, which was first shown by MENDEL and later in crossing experiments by OSTENFELD (1906, 1910) and embryologically by ROSENBERG (1907). The formation of embryos after the fertilization of legitimate embryo-sacs occurs also in a number of apomictic species of *Crepis* (STEBBINS and JENKINS, 1939) and even in species of *Rubus* (CRANE and THOMAS, 1940 a; LIDFORSS, see below). An entirely different type of *embryo formation* seems to occur in *Rubus*, viz. by the fusion of two nuclei from the embryo-sac (automixis, according to CRANE and THOMAS).

Origin of apomixis in Poa pratensis. — In *Poa pratensis* apomixis is also associated with polyploidy. The sexual types in the species do not, however, differ with respect to the absolute number of chromosomes from the apomictic types (Table 26). No definite connexion could be found between the degree of apomixis and the chromosome number. The haploid with $2n = \pm 36$ found by MÜNTZING was sexual, but it was nevertheless able to form embryo-sacs with an unreduced chromosome number. Strain 709, which had an exceedingly high chromosome number, $2n = \pm 90$, was also highly sexual. These two may serve as examples of the occurrence of sexuality.

The lowest euploid number found in the species is 28 (Table 28). It was found only once, and must therefore for the present be taken with a certain reservation. Solitary plants had ± 42, and $2n = 49$ seems to be the lowest euploid number from which plants with aneuploid numbers may in general have arisen. The chromosome numbers found vary so much between $2n = 49$ and $2n = 91$ that it cannot possibly be determined whether they are also derived from primary types with $2n = 56$, 63, etc. It is therefore difficult on the whole to decide which is the primary type. From Table 28, however, it is rather clear that $2n = 49$ should be a primary type. In view of the different ways described above in which aberrants can arise in a type with $2n = 49$, it is quite conceivable that all the chromosome numbers found were built up on the original number of 49. To what extent a *P. pratensis* plant with $2n = 49$ is an *autopolyploid* or *allopolyploid* is at present impossible to decide. Evidence in favour of a certain degree of autopolyploidy is afforded, on the one hand, by the occurrence

of types with 48, 50, 51, 52 chromosomes, etc., on the other hand, by the fact that gametes with widely varying chromosome numbers were found to be viable even at low 2n values (p. 60). In autopolyploids, as MÜNTZING (1937 b) has specially shown in *Dactylis* (also in *Allium*; LEVAN, 1936 a and b), the decrease or increase of a chromosome has no severe effect when the total chromosome number is high. As already pointed out by MÜNTZING (1933 a, 1940), and in accordance with the shown development of new forms, the aneuploid forms of *Poa pratensis* are to be regarded as true aneuploids and have probably arisen by the formation of fragments only to a small extent.

On the other hand, the low pollen-fertility and the weak seed setting in many plants from sexual families may however indicate allopolyploidy. The above-mentioned haploid plant with $2n = 36$ was thereto cytologically highly alloploid, which does not, however, exclude an autopolyploid constitution (v. MÜNTZING and PRAKKEN, 1940). By chromosome morphological studies FLOVIK (1938) considered himself in a position to infer allopolyploidy in a strain of f. compl. *alpigena*, »made up of two genomes from one of the parental species and four from the other». ARMSTRONG (1937) and BROWN (1939), too, regard *Poa pratensis* as an autoallopolyploid. The correctness of this view will probably be established by investigations of haploids. For the present we may safely say that the high chromosomal types may be autopolyploids of the low chromosomal types.

The genotypical basis of apomixis in *P. pratensis* has already been discussed a little by MÜNTZING (1940). MÜNTZING's assumption of a multifactorial basis for apomixis is entirely supported by material submitted. It should also be clear from the preceding discussion, according to which we should at least reckon with different factors that induce apospory and cause the autonomous parthenogenetic development of the aposporous embryo-sacs. That is also in complete agreement with GUSTAFSSON's statement (1935) that a distinction must be made between the origin of zygoid embryo-sacs and the stimulation, which causes the development of the egg-cell. Crossing experiments, too, furnish evident proof of a multifactorial structure. Otherwise it is difficult to understand why the aberrants between apomictic strains become sexual. If apomixis is the result of a multifactorial gene action it is therefore natural that the apomictic development is easily disturbed as soon as a change in the chromosome constitution, e. g. in aberrant formation, takes place. All tested aberrants were found to be highly sexual, even if the degree of sexuality probably varied. The observations

of the F_3 and the following generations have not yet been sufficiently extensive to elucidate this gradation. The hybrids with *Poa alpina* also behaved in the same manner as the pure *pratensis* aberrants. *P. alpina* is characterized by diplospory, and apospory does not seem to occur normally in the species. In an occasional hybrid a dominance for factors from *P. pratensis*, which induce apospory, was observed, aposporous cells being plainly discernible. But in these hybrids a parthenogenetic seed formation occurred, if at all, only to a small extent.

Aberrants arising in other apomictic species with apospory behaved somewhat differently as regards seed development. According to LIDFORSS (1905, 1907 a and b, 1914), in *Rubus* hybrids from species crosses between apomictic species give an intense segregation in F_2, but segregates from such species had apomictic propagation. Hybrids from crosses between species of *Hieracium pilosella* also behaved differently (OSTENFELD, 1910), while the hybrids of apomictic *Potentilla* obtained so far (A. and G. MÜNTZING, according to MÜNTZING, 1940) were constant in the following generation. Such differences should be expected according to the structure of the apomixis in different genera and the genotypic constitution of the crossed species.

But what has been said above explains only how apomixis may arise in *Poa pratensis* but not why apomixis is dominant in nature despite its undoubtedly being a very complicated product of segregation. In order to obtain a reply to the latter question comparisons between progenies of sexual plants and comparable apomictic strains with regard to vitality and fertility are especially valuable. MÜNTZING (1940) was able to make such a comparison in *Poa alpina* and he found that »the apomictic strains differ favourably and strikingly from sexual progenies by their uniformly good vigour and good seed production». Further, he says that »from the experience gathered in our material there is reason to believe that apomixis is an advantageous property, gradually evolved by natural selection».

These statements seem also to describe very closely the conditions in *Poa pratensis*. In preceding chapters comparisons were made, firstly, between the sexual strains 709 and 813 and their offspring with apomictic strains (p. 41), secondly, between aberrants in F_1 and F_2 and mother plants (pp. 75—78), and, thirdly, between sexual families obtained from plants in nature and apomictic strains raised in the same manner (p. 81). The families thus sexually produced (progenies of strains 709 and 813, aberrants and sexual plants found

in nature) showed a wide segregation in different properties. They differed somewhat with respect to fertility and vitality, but it was quite evident that they were, practically without exception, on the average inferior to the apomictic strains due to disturbances in at least some property important for vitality and fertility.

That was also the case with the diploid aberrants, which form a quite comparable material (p. 55) with the above-mentioned sexual families. Control tests were made in such properties as germinability, height, spreading capacity, yield, pollen formation, seed setting, etc. Among these families, however, there were fully vital and fully fertile plants. If these plants were able to unite apospory and parthenogenesis their progenies would be constant and on an average possess a greater ability to compete than the progenies of plants with sexual seed production. And it should be possible for a segregation of such plants to take place among the sexual families. An indisputable hereditarily conditioned tendency to apospory was shown in certain sexual families, and such a tendency should be expected in families obtained from aberrants from apomictic strains. There is also probably a similar tendency to a parthenogenetic development, although the material has not been studied long enough to establish that. Apospory and parthenogenesis can also be introduced into the sexual families by incrossing with apomictic plants. On account of their attributes, the apomictic strains thus produced, which may also be highly heterozygous, should be gradually rendered dominant by natural selection. The continual appearance of aberrants — which were sexual — makes this dominance incomplete. Nor is it inconceivable that certain sexual families possess such characters as enable them to survive with sexual seed production in nature at least for quite a long time. Such a family very probably occurred also among those investigated above. By means of the factors mentioned an equilibrium would arise in nature between the number of apomictic and sexual plants.

In apomictic material it has rather infrequently been possible to carry out investigations of fully comparable apomictic and sexual families in one and the same species as those of *Poa pratensis* and *alpina* dealt with above. Certainly several species are known with sexual and apomictic strains, e. g. *Crepis* (see above), *Festuca ovina* (LEVITSKY and KUSMINA, 1927; TURESSON, 1930, 1931), *Hieracium umbellatum* (ROSENBERG, 1927; BERGMAN, 1935 b), *Sorbus* (LILJEFORS, 1934), and *Taraxacum* (GUSTAFSSON, 1937), but in most cases the sexual and apomictic strains represented different and not quite comparable chro-

mosomal races. The results in *Poa pratensis* and *alpina*, however, resemble those obtained by LIDFORSS in his *Rubus* crosses. From LIDFORSS's Resumé (1914) it appears that in the F_2 of hybrids between predominantly apomictic *Rubus* species all transitions between complete sterility and normal fertility were obtained. In several cases a marked segregation was also obtained in the properties of vitality. Even the F_1 plants were often weaker than the parents, but they nevertheless often had good fertility. The segregation diminished, however, in the following generations, and LIDFORSS states that it is possible that an ultimately constant offspring may appear along these lines. Comparable are also OSTENFELD's above mentioned *Hieracium* crosses where he obtained in F_1, just as in *Poa*, segregation in vitality and fertility.

Thus from the material at present available it seems that apomixis in *Poa pratensis* and *alpina* arose as a complicated product of segregation, the frequency of which will be the result of a natural selection. In his review of earlier works on the origin of apomixis — particularly the hybridisation hypothesis according to WINGE (1917) and ERNST (1918) — SCHNARF (1929) expresses the view that in many cases the tendency to apomixis must have been present in the basic material of the apomictic species, and that this tendency was liberated but was not immediately evoked in the hybridisation. Apomixis would appear where the hybridisation disturbed the normal sexual propagation. Such an explanation has been accepted or put forward as being probable in many cases of apomixis (especially GUSTAFSSON, 1935, 1939 a and also, though more or less clearly, HOLMGREN, 1919; ROSENBERG, 1930; STEBBINS and JENKINS, 1939; DARLINGTON, 1939: apomixis, an escape from sterility). It appears as if *Poa pratensis* and *alpina* constitute two well-founded examples of the occurrence of apomixis in accordance with SCHNARF's hypothesis.

Polyploidy and polymorphism. — As mentioned above, the chromosome numbers found in *Poa pratensis* varied between $2n = 49$ and $2n = 91$, and *all* numbers between these two may occur. Chromosomal races are well known in the majority of apomicts, but the abundant occurrence of aneuploids seems to be mainly restricted to *Poa*. Some plants with 2n higher than 91 were obtained, and the highest value found so far in nature is $2n = \pm 124$. The chromosome optimum seems to lie between 95 and 100, and this limit is probably determined to a great extent by the plants, as a rule, becoming non-viable when the limit is overstepped. Such a limit has been shown for many plants (MÜNTZING, 1936, p. 286). It should be borne in mind that for an apomictic plant

with 2n = 100 the endosperm receives the chromosome number 250, provided that the central nucleus is fertilized. The occurrence of fertilized unreduced embryo-sacs has been shown even in plants with 2n = 90, therefore this *possibility* of an increase in the chromosome number exists also in the high chromosomal plants.

As pointed out by way of introduction, *P. pratensis* is exceedingly polymorphous, a characteristic it has in common with many apomicts. The problem concerning the origin of this polymorphism and the development of different forms is much simpler in the facultatively apomictic forms than in the total apomicts, even if the production of new forms is possible in the latter (DARLINGTON, 1937; GUSTAFSSON, 1934). By the formation of aberrants in apomictic strains, by the occurrence of sexual plants, the possibility of developing forms is intrinsically great in *P. pratensis*, and that furnishes a fully satisfactory explanation of the polymorphism of the species.

In the discussion on the triploid aberrants it was shown that in certain properties they exhibited changes which were probably due to the quantitative increase in the chromosome number. What are the facts then concerning such properties in the apomictic chromosomal races of *P. pratensis* found in nature? It has already been shown that the 1000-seed weight was highest in the high chromosomal strains, and frequently they also had a better seed setting. In the material planted out in 1936 a comparison was made between families with 2n = 50—60 and families with 2n = 80—90, i. e. in an increase of the chromosome number by about 50 per cent. Of the former 9 families, and of the latter 15 families were available. The properties compared were plant height, length of panicles, number of panicle branches at the basal node, average length and breadth of the two apical culm leaves and leaves of vegetative shoots. The average values obtained were as follows (Scheme 9; the probability of the difference being due to chance is also given).

At 2n = 60—70 and 70—80 the values for the properties mentioned, where marked differences occurred, lie mostly between those recorded in the scheme. Thus in the higher chromosome number the plant is shorter, has a shorter panicle, shorter and, in proportion to the length, broader culm leaves and also, although not so conspicuously, fewer panicle branches at the basal node. The sterile shoot-leaves are about equal as regards length and breadth.

The difference found in certain characters between high and low chromosomal strains thus resembles in many respects the difference

between the triploid aberrants and their mother plants, and that is especially true of the triploid aberrants from strain 704 (height, leaves and 1000-seed weight, see Table 20). A comparison with these alone is justifiable, partly because they have an approximately proportionate increase of chromosomes (53 → 80 in strain 704), partly because they were measured in exactly the same manner as the collected material. The change in leaf and 1000-seed weight from low to high chromosomal plants is also reminiscent of what was observed in autopolyploids (cf. MÜNTZING, 1936). STÄHLIN (1929) has shown that the maximal plant height seems to be attained by some grasses of the hexaploid—octoploid forms, but that it afterwards decreases. In conformity with that there is a very conspicuous decrease in height from 7-ploid to 13-ploid forms. Thus these comparisons do not seem to contradict our previous

Scheme 9. Morphology and chromosome number.

2n	Plant height cm.	Panicle length	Number of branches at basal node	Culm leaves			Sterile shoot-leaves	
				length cm.	breadth mm.	breadth / length	length cm.	breadth mm.
50—60	79,1	13,3	4,37	9,39	4,66	0,508	30,93	3,14
80—90	58,4	9,1	3,83	5,54	4,13	0,773	28,62	3,14
P	$< 0{,}001$	$< 0{,}001$	$< 0{,}05$	$< 0{,}001$	$> 0{,}05$	$< 0{,}001$	$> 0{,}05$	$> 0{,}05$

assumption that the high chromosomal strains, at least to a certain extent, have developed from the low chromosomal strains by a quantitative increase of the chromosome number.

In several cases an ecological or geographical differentiation has been observed in different chromosomal races of one and the same apomictic species, as in sexual species with different chromosomal races or in closely related species with different chromosome numbers (see MÜNTZING, 1936, 1937 b and TISCHLER, 1935). This problem has been studied very thoroughly in autopolyploids and allopolyploids of *Crepis* (BABCOCK and STEBBINS, 1938), in *Rubus* (GUSTAFSSON, 1939 b) and in *Festuca ovina* (TURESSON, 1930, 1931), the last-mentioned case of geographical segregation, however, has been criticised by FLOVIK (1938). Table 26 also conveys an impression of a certain geographical differentiation of chromosomal races. 15 of the 20 strains from Götaland and all the 5 strains from England were found to have a higher chromosome number than 80, while such a high number was found in only

1 of the 11 strains from Germany. 9 of these 11 German strains had less than 70 chromosomes. Of the 17 strains originating from the south-west coast of Sweden, some of them from Hallands Väderö, no less than 15 had $2n > 80$. This difference between the chromosome numbers of the strains from Germany and those of the strains from England and South Sweden can hardly be due to chance alone. Most of the material from Germany was collected in inland districts with little rainfall, while the material from South Sweden originates to a great extent from meadows along the shore. In visited parts of England *Poa pratensis* was very rare, and seemed to have been supplanted in the grasslands by other grass species, particularly aggressive in the maritime climate. The types found were short and often pruinous like the seaside type. It seems not improbable that they belong to one in the maritime climate selected type. Is that the case, the geographical differentiation mentioned may be an ecological differentiation. This differentiation seems to be particularly marked in the extreme chromosomal classes, e. g. $2n = 50—60$ and $2n = 80—90$. The material, however, is scanty in certain respects, and a further collection of seashore and arid soil types from other localities is necessary in order to clarify the ecological differentiation and to prove that differences in chromosome number are not associated with the phenomenon of immigration (cf. *Rubus*; GUSTAFSSON, 1939 b).

Further, somewhat different results were obtained in investigations of the problem under discussion by other workers. BROWN (1939), for instance, studies the connexion between the cytology and morphology of 10 different types with a 2n varying only between 42 and 64. He assumes $2n = 56$ to be the primary type, made up of 2 different genomes, 4 of each kind. These genomes are supposed to carry the genes for different morphological characters, and a certain morphological type arises depending on the number of genomes of the one kind or the other present. To obtain a connexion between chromosome number and morphology under such conditions it is also necessary to assume that low chromosomal plants have lost one type of genome and that the high chromosomal plants have increased the other type. No plausible reasons, however, can be put forward for these assumptions. It is clear, however, that BROWN's low chromosomal types were characterized by properties which were hardly to be expected from our results. TINNEY and AAMODT (1940) studied the question whether grazing or other environmental factors brought about a selection of a certain morphological type. Their results were negative. But they did not

investigate the chromosome number and therefore no idea can be formed as to what chromosomal races they worked with.

The form-complexes in Poa pratensis. — The present material does not permit any far-reaching conclusions as to the form-complexes, but on the other hand some elucidatory facts were obtained. It has already been pointed out in the introduction that the form-complex *alpigena* should be considered separately, and we shall therefore deal first with *eu-pratensis*, *angustifolia* and *irrigata*, but only the seashore forms of the last-mentioned, owing to the fact that the forest types (see p. 5) are scantily represented in the material. An account of the taxonomic difference was given on pages 4 and 5. The form-complexes, however, also grow in different habitats, a difference that is especially conspicuous in the case of *irrigata* and *angustifolia* (see, among others, HYLANDER in LAGERBERG's »Vilda växter i Norden», Part I, 1937, p. 163, where *angustifolia* is said to grow in arid soil and *irrigata* to be found in more or less damp soil). *Eu-pratensis* does not seem to be confined to any particular locality. No difference was found in the seed formation of the three form-complexes, and among them there were both apomictic and sexual forms. The form-complexes can be crossed with each other, but the F_1 as well as the F_2 showed not only a difference in chromosome number but also a difference in their propensities to different characters. In addition to strains 709 and 813, five very typical apomictic *irrigata* families were found. All five were obtained from the west coast of Skåne and they had, like strains 709 and 813, high chromosome numbers, $\pm$ 86, approximately 95, approximately 90, $\pm$ 87 and $\pm$ 84 respectively, and were different from each other. In five typical *angustifolia* strains, among them strain Ä 787, from Germany, and one such strain from Sweden, the following 2n values were obtained: $\pm$ 50, $\pm$ 50, $\pm$ 65, $\pm$ 64, $\pm$ 65 and $\pm$ 64, thus chromosome numbers clearly differing from those obtained in *irrigata*. These strains, two of which were sexual, also differed from each other morphologically, but they were of the same main type. *Eu-pratensis* appears also to be very labile as far as the chromosome numbers are concerned.

It is often asserted that *irrigata* and *angustifolia* are two originally wild complexes, whereas *eu-pratensis*, to a great extent at least, is derived from imported and cultivated *pratensis* (HYLANDER, l. c.). This does not seem improbable, and in view of what was said above we may venture to assume that *irrigata* and *angustifolia* are two wild complexes with different chromosome numbers, and are not only quantitatively but also qualitatively differentiated into several factors.

In that way these two complexes came to occupy different habitats. It remains, as pointed out in the preceding discussion, to throw further light on the extent to which the differentiation with respect to habitat is connected with the chromosome number. Further evidence in support of this connexion is found in the fact that we find high chromosomal *eu-pratensis* types in the shore formation. The *irrigata* and *angustifolia* types treated comprised part of the material from different regions, as mentioned above. But the material also contained *eu-pratensis*, probably selected, being possible by the presence of chromosomal races or by the formation of new such races (e. g. by the formation of triploid aberrants).

The form-complex *alpigena* has a geographically different distribution from those described above. It is a purely northern complex and is described as being very variable both with respect to characters and habitat. It has also been found to vary very much as regards chromosome number. The following 2n values were obtained: ± 46, ± 56, ± 66, ± 68, ± 77, ± 83 and ± 84. Thus this form-complex can hardly be compared directly with *irrigata* and *angustifolia*, possibly it is more comparable with *eu-pratensis*. It does not appear improbable that chromosomal races of *alpigena* with different ranges of distribution can be obtained, corresponding to *irrigata*, etc. A strain of *alpigena* was crossed with a strain of *irrigata* and gave a fertile F_1.

The form-complexes in *Poa pratensis* can thus be crossed with each other, and quite certainly crosses occur in nature where the complexes meet, for instance, through the sexual types. In this way transitional forms arise, which are also well known from nature. The investigations therefore completely confirm NANNFELDT's statement, quoted in the introduction, that *Poa pratensis* L. (coll.) should be regarded as a very polymorphous species (an agamic heteroploid complex, according to BABCOCK and STEBBINS, 1938) and that single specimens deviating in one or other character cannot possibly be described as »new species».

SUMMARY.

1. In *Poa pratensis* L. (coll.) there are forms with mainly apomictic seed production, other forms with mainly sexual seed production as well as transitions between these two forms. The apomictic types are predominant in nature. No evident connexion was found between chromosome number and seed formation (apomictic or sexual).

2. Pollen formation in the apomictic strains, which are pseudogamous, is on the average good. A few exceptions exist. The good pollen formation is probably to a certain extent the effect of selection, for a greatly reduced pollen fertility frequently occurs in the progeny of sexual plants.

3. In the apomictic strains we meet with aposporous gametophyte development and usually autonomous parthenogenesis with a stimulation (most probably fertilization) of the central nucleus. The aposporous E.S. may be fertilized. Haploid, diploid and triploid aberrants have been obtained from apomictic strains.

4. Independent of the mode of origin of the aberrants, their progenies are more or less variable, indicating a mainly sexual seed production in the aberrants.

5. Apomixis is considered to be due to many factors, apospory and parthenogenesis being conditioned by different factors. The predominant occurrence of apomixis is explained as the result of selection, which was proved by a comparison of the vitality and the fertility in apomictic and sexual families.

6. The lowest 2n found in the species is 28 and the highest $\pm$ 124. Values below 49 and above 91 are exceptional. All values between $2n = 49$ and $2n = 91$ seem to be equally frequent. In view of the manner in which aberrants are formed, the chromosome numbers above 49 can be explained as having arisen from a primary type with $2n = 49$. Other primary types may have occurred however.

7. The polymorphism of the species appears to be in conformity with the continual formation of new forms, which must take place in the aberrant-formation and through the sexual families. Different chromosomal races can be crossed with each other. The relationship between chromosome number, on the one hand, morphological characters, geographical and ecological distribution, on the other hand, is discussed. Differences in geographical distribution have been shown. This is possibly a question of ecological differentiation.

8. Apomictic and sexual seed formation has been shown in the form-complexes *eu-pratensis*, *irrigata*, *angustifolia* and *alpigena* set up by taxonomists, and occasionally designated as species. (No predominantly sexual type has so far been found in *alpigena*.) These form-complexes can be crossed with one another. There is a possibility that *irrigata* (the shore form) and *angustifolia* are two wild chromosomal races, also differentiated by specific genes. *Eu-pratensis* and

alpigena are highly variable with respect to chromosome number, and they may not be quite comparable with the former two.

9. Several morphologically widely different hybrid plants between *Poa pratensis* ♀ and *Poa alpina* ♂ have been obtained. They arose partly by the fertilization of reduced, partly by the fertilization of unreduced E.S:s of *pratensis*. They were all non-viviparous. The F_1 had a highly sexual seed production, even after crossing with apomictic races of *pratensis* and *alpina*. In one or two F_1 plants we came across reduction division (probably greatly disturbed) with megaspore formation and even aposporous E.S. initials (*pratensis* has apospory, *alpina* diplospory).

10. The following chromosome numbers were found in *Poa* species (in addition to *pratensis*): $2n = \pm$ *alpina* 31, *bulbosa* 28, 45, *trivialis* 14 + one trisomic plant ($2n = 15$), *compressa* 35, 42, 49, *nemoralis* 28, 42, *glauca* 65. A hybrid plant was obtained from the cross *pratensis* × *glauca*.

Lännäs, Undrom, April 1941.

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CONTENTS.

I. Introduction 1

II. Experimental 3

1. Investigations of 12 specially selected strains 3
A. Taxonomy 3
B. The morphology of the 12 strains 6
C. Somatic chromosome numbers 9
D. Development of the male gametophyte 11
E. Development of the female gametophyte 16
F. Comparison between the development of P.M.C:s and E.M.C:s in strains 709, 703 and 746. 24
G. Pollination experiments 25
H. Seed setting 28
I. 1000-seed weight 35
J. Germination tests 36
K. Polyembryony 39
L. Progeny generations of the sexual strains 709 and 813 41
M. Progeny generations of the apomictic strains Ä 787, 701—704, 5303, 746, G 258, 768 and G 125 46
N. The morphology, etc. of haploid, diploid and triploid aberrants .. 53
O. F_2 and F_3 of diploid and triploid aberrants 61

2. Progeny investigations of material collected in nature 78
A. Introductory notes 78
B. The occurrence of apomictic and sexual seed formation 79
C. Morphology, chromosome number, etc. of the families showing a high degree of sexual seed formation 81
D. Morphology, chromosome number, etc. of the families with a high degree of apomictic seed formation 85

3. Results from crosses with *Poa alpina* 91
A. Introductory notes 91
B. F_1 plants of *Poa pratensis* × *alpina* 94
C. F_2 and back-crosses families 99
D. Some conclusions 103

III. Discussion 105

Summary 120

Literature cited 122

THE INDIVIDUALITY OF SOWS IN REGARD TO SIZE OF LITTERS

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IN order to judge the selectional value of such characters as litter size and litter weight of sows it is important to know if different sows at repeated farrowings show any marked individuality in this respect. It is also of importance to learn if the individuality may be more or less strongly marked at different ages of the litters. It seems that the stage of greatest individuality should be the best moment to obtain succesful selection. LUSH and MOLLN (1937) have studied some of these characters by means of analysis of variance, and they use the intraclass-correlation as a measure of the differentiation between individual sows. In the present study the numbers of pigs per litter for a number of sows have been analysed and the coefficient of the intraclass-correlation has been determined with the usual formula

$$\frac{\text{total variance—variance within sows}}{\text{total variance}}.$$

THE INDIVIDUALITY IN REGARD TO NUMBER OF PIGS AT FARROWING.

The material used was obtained from the results of farrowings in the Wiad herd. All farrowings from sows giving at least 3 litters are included. The number of sows is 117 with 596 litters, on an average 5,1 litters per sow. The total number of pigs, dead or alive at farrowing, is 6333 or 10,6 pigs per litter. The analysis of variance of the numbers of pigs per litter at farrowing is recorded in Table 1.

In the material of LUSH and MOLLN there were differences between herds, a cause of variation which is absent in the present material. Our data have not been corrected for age of sow, but this cause of variation is at least partially eliminated by grouping the sows according to the number of their litters.

A simple comparison between *inter*-sow variance and total variance (Table 1 a) gives a coefficient of 0,29. If the sows are grouped, as done by LUSH and MOLLN, according to their birth-year in contemporary

sows (Table 1 b) the same coefficient is obtained. In the present material the year of birth of the sow is of no importance and has, consequently, been disregarded in the following analyses.

TABLE 1. *Analysis of variance of number of pigs farrowed per litter.*

	Degrees of freedom	Sum of squares	Mean square	Variance ratio	Intraclass-coefficient
a)					
Total	595	5313,57	8,93		
Between sows...	116	2271,48	19,58	$\frac{19,58}{6,35} = 3,08$ [1]	$\frac{8,93 - 6,35}{8,93} = 0,29$
Within sows.....	479	3042,09	6,35		
b)					
Total	595	5313,57	8,93		
Between groups of contemporary sows	6	61,09	10,18	$\frac{20,09}{10,18} = 1,97$ [4]	$\frac{8,93 - 8,92}{8,93} = 0,001$
Within groups of contemporary sows	589	5252,48	8,92		
Between sows	110	2209,95	20,09	$\frac{20,09}{6,35} = 3,16$ [1]	$\frac{8,92 - 6,35}{8,92} = 0,29$
Within sows...	479	3042,53	6,35		
c)					
Total	585 [5]	4944,86	8,45		
Between breeds	1	45,31	45,31	$\frac{45,31}{8,39} = 5,40$ [3]	$\frac{8,45 - 8,39}{8,45} = 0,008$
Within breeds...	584	4899,55	8,39		
Between groups of sows with equal number of litters	12	538,60	44,88	$\frac{44,88}{7,62} = 5,89$ [1]	$\frac{8,39 - 7,62}{8,39} = 0,09$
Within groups of sows with equal number of litters	572	4360,95	7,62		
Between sows	102	1501,48	14,72	$\frac{14,72}{6,08} = 2,42$ [1]	$\frac{7,62 - 6,08}{7,62} = 0,20$
Within sows...	470	2859,47	6,08		

The significance is denoted thus: [1] $P < 0,001$; [2] $0,001 < P < 0,01$; [3] $0,01 < P < 0,05$; [4] $P > 0,05$, where P is the probability that only random variation occurs. [5] In the group with 10 litters there was one sow only and therefore omitted in this case.

In Table 1 c the material has been grouped according to breeds (Large White and Improved Swedish Landrace) and number of litters

per sow. This number has varied from 3 to 10 and the number of sows with different numbers of litters is as follows:

Number of litters ...	3	4	5	6	7	8	9	10	Total sows	Total litters
Number of sows with different numbers of litters	28	26	18	17	15	7	5	1	117	596

The grouping according to number of litters must be important. For sows with a low number of litters only the first litters are recorded and it is known that these have a lower number of pigs than the later ones. Furthermore, sows with markedly low numbers of pigs in their first litters are often slaughtered, and in the above material the poorest sows are, therefore, to be found in the classes with only few litters. It is therefore to be expected that the lowest numbers of pigs per litter should be found among the sows with small numbers of litters.

Table 1 c shows that in the present material the difference between breeds is statistically insignificant and has but very slight influence on the coefficient of correlation. The number of litters per sow, as expected, is of considerable importance. If breed and number of litters are simultaneously considered, the coefficient is only 0,20, which value, however, is considerably higher than that obtained by LUSH and MOLLN.

TABLE 2. *Number of pigs at different ages of the litters.*

	Total	% of farrowed	Average number per litter
Farrowing, including dead-born	5060	100,0	11,0
» , alive	4670	92,3	10,1
Alive after 1 week	4001	79,1	8,7
» » 2 weeks	3893	76,9	8,4
» » 3 »	3851	76,1	8,4
» » 4 »	3723	73,6	8,1
» » 5 »	3658	72,3	7,9
» » 6 »	3585	70,8	7,8

THE INDIVIDUALITY IN REGARD TO SIZE OF LITTER MEASURED AT DIFFERENT AGES OF THE LITTERS.

It is of interest to know at which age of the pigs the size of the litters should be determined so as to show most clearly the differentiation

between the sows. In a study of this problem we have utilized all sows having at least 3 litters with all pigs unweaned at the age of six weeks. The material analysed has thus been limited to 97 sows with 461 litters. For each of these litters we know the total number of pigs at farrowing, the number of pigs alive at farrowing and the number of pigs alive at the end of 1, 2, 3, 4, 5, and 6 weeks after farrowing.

TABLE 3. *Analysis of variance of numbers of pigs per litter at different ages of the litters.*

Age of litters		Total	Between sows	Within sows	Variance ratio	Intra-class-coeffi-cient
		460	96	364		
Farrowing(in-cluding dead-born)	Sum of squares	4468,043	1725,891	2742,152	$\frac{17,98}{7,53} = 2,39$ [1]	0,22
	Mean square	9,71	17,98	7,53		
Farrowing (alive)	Sum of squares	3468,191	1348,881	2119,310	$\frac{14,05}{5,82} = 2,41$ [1]	0,23
	Mean square	7,54	14,05	5,82		
1 week	Sum of squares	2690,444	989,147	1701,297	$\frac{10,30}{4,67} = 2,21$ [1]	0,20
	Mean square	5,85	10,30	4,67		
2 weeks	Sum of squares	2642,831	936,863	1705,968	$\frac{9,76}{4,69} = 2,08$ [1]	0,18
	Mean square	5,75	9,76	4,69		
3 weeks	Sum of squares	2631,367	914,366	1717,001	$\frac{9,52}{4,72} = 2,02$ [1]	0,18
	Mean square	5,72	9,52	4,72		
4 weeks	Sum of squares	2561,343	791,086	1770,257	$\frac{8,24}{4,86} = 1,70$ [1]	0,13
	Mean square	5,57	8,24	4,86		
5 weeks	Sum of squares	2524,048	739,467	1784,581	$\frac{7,70}{4,90} = 1,57$ [2]	0,11
	Mean square	5,49	7,70	4,90		
6 weeks	Sum of squares	2591,987	746,604	1845,383	$\frac{7,78}{5,07} = 1,53$ [2]	0,10
	Mean square	5,63	7,78	5,07		

[1], [2] as in Table 1.

The numbers of pigs at each time are given in Table 2. For each of these eight sets of data the coefficient of intraclass-correlation has been calculated (Table 3). The sole object of this study was to determine the relative sizes of coefficients, determined at different ages of the litters, therefore variations in breed and in number of litters per sow have been disregarded.

It is evident from Table 3 that the coefficient tends to decrease with increasing age of the litter, a phenomenon which was to be expected. Though the sows differ in milk production and care of their offspring, the environment increases in importance with the ageing of the litters and concomitantly the differences between the sows decrease.

The coefficient of correlation, as determined on the number of pigs at farrowing, is smaller in this material than in that presented in Table 1, although the difference is hardly significant.

TESTING THE INDIVIDUALITY BY SELECTION.

THE PRACTICAL RESULTS OF SELECTION.

The analysis of variance of the present material gave a coefficient of intraclass-correlation of $+ 0{,}20$. In order to test this figure and to demonstrate its significance in practical breeding work the following calculations were made: The sows have been grouped according to the number of pigs in their first litter. The total average of this number is 9,3. In group A are placed all sows with 9 pigs or less in their first litter, in group B those with 10 or more. In group A there are 56 sows with an average number of 6,8 pigs, in group B 61 sows show an average of 11,6. The difference between the two averages is 4,8 pigs. The coefficient of intraclass-correlation being 0,20, it is to be expected that 20 % of this difference will remain between the two groups, if the average number of pigs in the subsequent litters is determined. This expected difference is $\frac{20 \times 4{,}8}{100} = 0{,}96$. The empirical data are as follows: The sows in group A have given 220 later litters with an average of 10,4 pigs per litter. The sows of group B have 11,4 as an average of 259 later litters. The difference 1,0 fits the expected difference of 0,96 remarkably well.

If no selection is made the average number of pigs in the later litters is 10,9. The rejection of all sows with a number of pigs below the average in the first litter increases the average number of pigs in the subsequent litters by 0,5 or somewhat less than 5 %.

The difference between individual sows is most pronounced when determined on the number of pigs at farrowing and is considerably less if determined on the number alive six weeks later. From the pig-breeder's point of view, however, the selection of mother sows should be as efficient as possible with respect to the economical value of the litter at the time of weaning or at some fixed age near this time. The number of pigs alive six weeks after the first farrowing may be more closely correlated with the number and weight of the subsequent litters at weaning than the number of pigs at birth in the first litter. The former number may, therefore, be a better basis of selection, in spite of the fact that it offers a poorer measure of the differentiation between sows. It is also quite possible that the selection will be most effective if it is based on both numbers: pigs at the first farrowing and pigs alive six weeks later.

In order to test the effects of the different modes of selection the following calculations were made: The 97 sows have been divided into four groups according to the numbers at birth of the first litter and the numbers alive six weeks later. The four groups are:

9 : 7. 9 pigs or less at birth, 7 or less alive after six weeks.
9 : 8. Same numbers at birth, 8 or 9 alive after six weeks.
10 : 7. 10 pigs or more at birth, 7 or less alive after six weeks.
10 : 8. Same numbers at birth, at least 8 alive after six weeks.

With this grouping it was possible to study the effects of different modes of selection of mother sows after the results of the first farrowing. Table 4 shows the results of three different modes of selection, calculated with respect to number of pigs at weaning as well as with respect to total weight of litter at this time.

The three »selection experiments» carried out on the material are as follows:

(1) The 97 sows are grouped according to the number of pigs at birth in the first farrowing.

A: 9 pigs or less, 9 : 7 + 9 : 8.
B: 10 pigs or more, 10 : 7 + 10 : 8.

The average number of pigs per litter at weaning and the mean weight of litter at the same time are determined for all subsequent litters of sows in group B.

(2) The sows are grouped according to the number of pigs in the first litter alive after six weeks.

A: 7 pigs or less, $9 : 7 + 10 : 7$.
B: 8 pigs or more, $9 : 8 + 10 : 8$.

The average result of the subsequent litters of group B is determined as in (1).

(3) The sows are grouped according to numbers at birth and six weeks later.

A: 9 pigs or less at farrowing and 7 or less six weeks later, $9 : 7 + 9 : 8 + 10 : 7$.

B: 10 pigs or more at farrowing and 8 or more at six weeks, $10 : 8$.

The average results of all subsequent litters of group B are determined as in (1) and (2).

The results of the 3 different modes of selection are compared with the average result of all subsequent litters from all 97 sows, i. e. the whole material without selection. The corresponding data are given in Table 4 as selection number 0.

It is evident from Table 4 that the number of pigs alive 6 weeks after the first farrowing is of more value in practical breeding than the number born in the first litter. The rejection of all sows below the average in the latter number only increases the result in the subsequent litters at weaning by 2,8 % in number and 1,4 % in weight, whereas the rejection according to number alive after six weeks has an improving effect of 4,4 % and 3,4 %, respectively.

The best result is obtained if the selection is made as in (3). In this case all sows are eliminated which have a fecundity below the average in their first litter and also all sows with good fecundity but poor capacity for maintaining their offspring until weaning age. By this selection an increase of 5.8 % is gained in number and of 3,6 % in weight at weaning of the subsequent litters.

It is of interest to note that the selection according to number has a smaller effect on the weight of litter than on the number. Environmental factors must have a greater influence on weight than on number, and that was to be expected.

According to the data of the present paper the individuality of sows in producing litter of a certain size is more pronounced than in the material of LUSH and MOLLN. There seems to exist some possibilities of making a selection based on the number of pigs in the first litters of the sows. This conclusion is also supported by the results of certain German investigations (HARING und HAGEN, 1939). Studies of

TABLE 4. *Number of pigs and litter weight at 6 weeks in the later litters of the sows if selection is made according to pig number in the first litter.*

Selection	Group B		Group A		Number of pigs at 6 weeks in later litters		Litter weight at 6 weeks in later litters	
	Selected sows		Rejected sows					
	Group	Number of sows	Group	Number of sows	Per litter	%	Kg. per litter	%
0	10:8 + 10:7 + 9:8 + 9:7	97	— — — — — — — —	0	7,77	100,0	81,4	100,0
1	10:8 + 10:7	53	9:8 + 9:7	44	7,99	102,8	82,5	101,4
2	10:8 + 9:8	51	10:7 + 9:7	46	8,11	104,4	84,1	103,4
3	10:8	42	10:7 + 9:8 + 9:7	55	8,22	105,8	84,3	103,6

0 = no selection; 1 = selection on pig number at farrowing; 2 = at 6 weeks; 3 = combined selection at farrowing and 6 weeks.

the same kind but bearing on the individuality of the weights of pigs and litters are now in progress.

SUMMARY.

The results of 596 farrowings of 117 sows of the Wiad herd were studied, all sows giving at least 3 litters.

The birth-year of the sow had no influence on the number of pigs per litter. There was a small and insignificant difference in this respect between the two breeds studied, viz. Large White and Improved Swedish Landrace.

If the material is grouped according to breed and number of litters per sow there is an intraclass-correlation of $+ 0{,}20$.

461 litters from 97 of the sows were kept together at six weeks. On this material the coefficients of intraclass-correlation was determined on the following 8 sets of data: number of pigs at birth; number of pigs alive at birth; number of pigs alive at the end of 1, 2, 3, 4, 5 and 6 weeks after birth. The value of the coefficient decreases with increasing age of the litters.

If all sows are divided into two groups, above and below the average with respect to number of pigs born in the first litter, the difference in this number between the groups is 4,8. As the coefficient of intraclass-correlation is 0,20, it was to be expected that there should be a difference of 0,96 between the averages of the two groups in regard to all subsequent litters. The actual difference observed is 1,0, and by rejection of all sows below the average in the first litter the result in the subsequent litters is improved by 5 %.

The sows were grouped according to number born in the first litter and number alive six weeks later, and the effect of selection, according to these numbers, on the average number and weight of all subsequent litters at weaning was studied. The selection according to number alive after six weeks proved to be more effective than the selection according to number born. The best result of the selection was obtained when due regard was paid to both these numbers.

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LINKAGE STUDIES IN PISUM SATIVUM × P. ABYSSINICUM

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(A Preliminary Report)

IN the winter of 1933 a series of crosses were started at the Genetic Institute of Lund University, Svalöf, between some thirty strains representing different gene combinations of *Pisum sativum* L. and *P. abyssinicum* BRAUN, a type differing widely morphologically from the former, and is generally regarded as a separate species (BRAUN, 1841; BOISSIER, 1872; JACKSON, 1895). Most of these crosses have been repeated in different years, and have lately been increased so as to include a number of other species of the genus, viz. *P. elatius, thibetanicum, asiaticum, fulvum* and *humile*. In 1937 the experiments were transferred to the S. S. A.'s Beet Breeding Station Hilleshög, Landskrona, where they have been carried on since then.

The great difference, genetically and physiologically, between *P. sativum* and *P. abyssinicum* is plainly evident in the experiments. A few instances may be given here. The successful crosses were few in number, and the seed setting of these crosses was low (0,354 seeds per pollination). The determination of the pollen fertility in F_1 shows an average of 37 per cent good pollen, but some plants occur that are, practically speaking, entirely fertile, and others again that are completely sterile. In normal self-fertilisation the seed setting in the F_1 and F_2 is most frequently about 1 seed per pod. In the F_2 in the majority of crosses there appear, firstly, several kinds of chlorophyll types, which soon die off, secondly, relatively rare »rogue» plants that frequently reach the flowering stage but are completely sterile. *P. abyssinicum* itself flowers 5—10 days earlier than low, white *sativum* types, and transgressions have occurred in certain crosses.

The purpose of the investigation was in the first place to study the linkage values in *P. abyssinicum* and the relation of the species to *P. sativum*, and subsequently also the position of other species in the genus. As *P. sativum* is one of the most thoroughly investigated plant species with respect to linkage, it is of importance to know the dis-

tribution of the genes also in other species of the genus. Species crosses in *Pisum* comprising the species *humile* and *thibetanicum* and a species from the Caucasus have already been investigated by HÅKANSSON (1936), LUTKOV (1930), PELLEW and SANSOME (1931), SANSOME (1932, 1939) and Miss SUTTON (1937) mainly from the cytological point of view. Genetical data have been published only by FEDOTOV (1930) and PELLEW (1931).

In his crossing series the author has in principle applied the following methods of investigation: the F_1 was analysed as far as possible with respect to pollen fertility and cytology, the F_2 with respect to segregation of factors and linkage, F_3 and further generations were sown as required, difficulties being encountered, however, owing to the invariable occurrence of partial sterility. Flowering time and the number of seeds per pod were determined in all generations. Attention was paid to the difference between reciprocal crosses and the influence of the different years on the results.

In spite of the marked disturbances incident to the crosses as a whole and also the segregations of the individual genes, the divergencies are nevertheless not greater than that the monohybrid segregations correspond with the 3 : 1 ratio previously known in *P. sativum*, and that the interaction between them could be employed to extend our knowledge of linkages in *Pisum*.

The following 30 genes have participated in the crosses: *Le*, *La*, *Lb*, *Td*, *D*, *Gp*, P_1, *A*, *Ar*, *B*, *Am*, Wb_2, *Fa*, *St*, *K*, *Tl*, *P*, *V*, *Btb*, Cp_1, Cp_2, *S*, *M*, *F*, *I*, R_1, (R_2?), *Pl*, U_1 and U_2. [For the designations of the genes the reader is referred to the works of WELLENSIEK (1925), MATSUURA (1933) and WINGE (1936)].

As mentioned above, the monohybrid segregations differ widely from the expected ratios. Some extreme examples may be given to illustrate this: $Tl : tl$ gave the ratio $3{,}45 : 0{,}55 \pm 0{,}031$, $B : b$ the ratio $3{,}38 : 0{,}62 \pm 0{,}068$, while $St : st$ and $R : r$ gave $3{,}29 : 0{,}71 \pm 0{,}061$ and $3{,}72 : 0{,}28 \pm 0{,}030$ respectively. Recounted according to the 15 : 1 ratio, the last-mentioned segregation gives a ratio of $14{,}86 : 1{,}14 \pm 0{,}068$, which indicates the presence of two cumulative factors in the segregation. But at the present stage of the investigation it cannot be decided whether this interpretation is correct, i. e. that *P. abyssinicum* is $R_1\ R_2$, or whether the influence the thick and hard seed coat of *abyssinicum* may have on the reliability of an ocular determination of the character of the seed furnishes an explanation of the deviation from the 3 : 1 segregation.

HADFIELD and CALDER (1935) seem to have obtained a similar distribution of the *R* factors in crosses between a few strains of *P. sativum*. The author's R_1 and R_2 correspond to their *R* and L_1 respectively.

The appearance of less viable types is a common occurrence in species crosses. A large number of such types was also met with in this investigation, partly in the form of poor germination in F_1 and F_2, partly as semi-vital plants (e. g. chlorophyll types). Some of the results of the present crosses, as for instance in a number of monohybrid segregations, also point to a connexion between plant deficiency and certain recessive genes. Further, it may be mentioned that the »chenille» character (*S*) proved almost impossible to determine, owing to the partial sterility usually occurring in the progeny, which directly inhibits the manifestation of the »chenille» character. One of the author's crosses furnishes further confirmation of RASMUSSON's (1927) division of the *Cp* factor (straight—bent pods) into Cp_1 and Cp_2 ($Cp : cp$ $15{,}_{11} : 0{,}_{89} \pm 0{,}_{34}$), but our segregation ratios indicate the presence of two cumulative genes.

Some new genes have been investigated. Thus, the indentation of the leaflets described by SUTTON (1911, 1914) was found to be a single recessive character ($Td : td = 2{,}_{95} : 1{,}_{05} \pm 0{,}_{015}$). The dominance, however, does not seem to be complete. The violet colouring of the seed coat (»*obscuratum*» seed) mentioned by VILMORIN (1911) is interpreted by KAJANUS (1913) as a modification of *F* (violet dots on the seed coat). In some of our crosses in which the parents were *F*, i. e. showed these dots, there was a segregation of the *obscuratum* character. It appears to be controlled by two complementary genes (U_1 and U_2), but is also influenced by a number of modifiers. If we contrast the stout, robust type of development of *P. sativum* and the thin, slender *P. abyssinicum*, we shall obtain a numerical proportion indicating that two complementary genes control the general difference in the mode of growth between them, with *sativum* as the dominating type. An unvarying characteristic of *P. abyssinicum* is a branchiness at the base of the plant (excluding those cases in which mechanical resistance may have caused ramification in the formation of shoots). *P. sativum*, on the other hand, is seldom ramose. It seems as if *P. abyssinicum* is recessive for two or three basic genes as regards this character.

Having regard to the 30 factors investigated, the gene formula of *Pisum abyssinicum* seems to be: *Le*, *La*, *lb* or *la*, *Lb*, *td*, *d*, *Gp*, p_1, *A*, *Ar*, *B*, *Am*, Wb_2, *Fa*, *St*, *K*, *Tl*, *P*, *V*, *Btb*, Cp_1, Cp_2, *S*, *m*, *F*, *I*, R_1, (R_2?), *Pl*, U_1, U_2.

As mentioned above, the linkages in these series of crosses could be studied to their full extent, and the correctness of the values was established still more firmly by comparing the crossing-over values obtained by different methods of determination, therefore the linkage data submitted can be safely employed in studying the arrangement of the genes in the genus *Pisum*.

Altogether 142 interactions between the 30 genes participating in the segregations have been investigated, 62 of which have not been previously reported. Referring the reader to the lists made up by WELLENSIEK (1925), MATSUURA (1933) and WINGE (1936), we shall give here only the arrangement into chromosome groups and the sequence of the genes in these chromosomes based on the collocation of the new data obtained in the present investigation and the findings reported in the above-mentioned earlier works.

Chromosome I (*D—P_2—O—I—Re* . . (1) . . *Fa—Z* . . (2) . . *Uni—M—Mp—F—B—Am—P_1—St* . . (3) . . *S—Wb_2—K*). — The dots denote the places at which the coupling is less sure, and these places are numbered (1), (2) and (3). As regards (1) only two linkage values are available, viz. *Re—Fa* = 42 % and *I—Fa* = 44 % (WINGE, 1936). At point (2) the relation *Z—Uni* has not been investigated and the linkage *Z—M* shows co = 42 % (WINGE, 1936). The combination of the group on the extreme right (3) *S—Wb_2—K* with this chromosome is not so probable. It is true that the author found a weak linkage between *St—Wb_2* (43 %), *B—K* (38 %), *B—Wb_2* (44 %) and *F—Wb_2* (45 %), but, on the other hand, a free combination between *St—K*, *F—K*, and SVERDRUP (1927) between *P_1—K*. However, if we let *K* and *S Wb_2* (the co-value of these genes is only 1—5 %) change places the assigning of *S—Wb_2—K* in this chromosome will be more probable. According to the results available, the gene *Am* (pink-white flower colour; DE HAAN, 1930) has its place between *B* and *P_1*, but it can also be placed between *F* and *B*.

Chromosome II (*Bta—Le—V—P—Wlo—Pl—Fl* . . . . *U*). — The gene *V* has been placed to the left of *Le* by other authors (RASMUSSON, 1927), but our results indicate that it should be placed to the right of *Le*, between that gene and *P*. *Wlo* (E. NILSSON, 1933, 1939) was placed between *P* and *Pl* on the basis of NILSSON's data. One of the *U* factors may have its place on the extreme right in the same chromosome. A suggestion of linkage between *Le* and *R* (co = 45 %) furnishes further evidence of the above-mentioned division of the *R* factor, because this

linkage assigns *R* to chromosome II, although other known linkage values place *R* in chromosome III.

Chromosome III (*A—Fta*(*Lf*)*—Tl—R—Pa—Q—Cp*$_1$*—N—Btb*). — On the basis of E. NILSSON's (1939) findings and the results of the present investigation it has been possible to combine the smaller gene groups *A—Fta* (RASMUSSON, 1935) with the larger group *Pa—N—Cp*$_1$*—Q—Btb*. It was found that genes *N* and *Q* should change places.

Chromosome IV (*Oh—Ar—Bl*). — This group has remained unchanged since WINGE's classification in 1936.

Chromosome V (*Fs—Gp*). — The group was set up by WINGE in 1936 (co = 42 %), because he found different linkage relationships for the character violet spots on the seed-coat from those previously known for *F*. WINGE introduced a new factor (*Fs*), which was said to be linked to *Gp*. Our findings agree essentially with older results, and thus the gene for violet spots in *P. abyssinicum* should be identical with the *F* factor of TEDIN and TEDIN (1928) and WELLENSIEK (1927, 1929, 1930). But *F* showed some divergent instances of linkage also in the present material, viz. *F—V* (23 %) and *F—P* (31 %), which should assign *F* to chromosome II. Further investigations are necessary to clarify these linkages. But two facts diminishing the value of the linkage determinations may be mentioned here. One is the variation in the manifestation of the violet dots, which occasionally renders the evaluation difficult, as also pointed out by WINGE (1936), the other is the occurrence of the *U* genes (a deep violet colouring of a larger or smaller extent of the seed-coat) in all crosses, because the *U* character is epistatic to *F*.

Chromosome VI (*La* or *Lb—Td*). — In our material this linkage is suggested by co = 42 %, but it is not statistically significant.

Summarizingly it may be said that the genes in *Pisum sativum* and *P. abyssinicum* appear in general to be homologous and that their reciprocal arrangement on the chromosomes seem, generally speaking, to be the same. There appears, however, to be a tendency to lower linkage values in a cross between these two species.

Hilleshög, April 1941.

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DIE KOPPELUNGSGRUPPE Gp—Cp—Fs—Ast VON PISUM

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(With a Summary in English)

EINLEITUNG.

DIE vorliegende Arbeit soll die Koppelungsverhältnisse folgender vier Eigenschaftspaare bei *Pisum sativum* behandeln: gerade—gekrümmte Hülse, grüne—gelbe Hülsenfarbe, punktierte—nicht punktierte sowie gestreifte—nicht gestreifte Samenschale. Bisher war über die diesen Eigenschaften zugrunde liegenden Gene und ihre Beziehungen zueinander und zu anderen folgendes bekannt.

Der Unterschied gerade—gekrümmte Hülse wird in der Hauptsache durch ein einziges Gen bedingt. Dieses wurde von WHITE (1925) mit dem Symbol *S* und von WELLENSIEK (1925 a) etwa einen Monat später mit *Cp* bezeichnet. Bald darauf hoben WELLENSIEK (1925 b) und vor kurzem E. NILSSON (1939) hervor, dass WHITEs Symbol S_s die Priorität zukommt und daher beizubehalten wäre. Dieser Ansicht kann jedoch nicht beigepflichtet werden, da das Symbol *S* schon viel früher (WHITE, 1917) für ein Gen verwendet worden ist, das in seiner rezessiven Form Verklebung der reifen Samen miteinander bedingt (sog. Chenille-Typus). Und die Benutzung desselben Symbols für ein anderes Gen, aber mit einem Index (einem Subskript) ist nicht gestattet; Indices sind nur für verschiedene Allele von multiplen Genen zu benutzen. S_s ist daher zu streichen. Von den meisten Forschern ist auch das Symbol *Cp* verwendet worden. — Die Spaltung im Eigenschaftspaar gerade—gekrümmte Hülse ist in der Mehrzahl der Kreuzungen gewöhnlich nicht rein, d. h. es treten Typen auf, die eine schwächere Krümmung der Hülsen zeigen, und bezüglich der man im Zweifel ist, ob sie als gerade oder gekrümmt zu klassifizieren sind. Man erhält aber auch keine Spaltung nach 1 *Cp* : 2 *Cpcp* : 1 *cp*. In gewissen Kreuzungen findet man jedoch, wie später gezeigt werden soll, klare Spaltung nach 3 *Cp* : 1 *cp*. Das Auftreten von Zwischentypen mit schwacher Krümmung dürfte daher wahrscheinlich auf Beeinflussung durch andere Gene zurückzuführen sein.

Dass die grüne Hülsenfarbe über die rein gelbe vollkommen dominiert und dass diese Farben in Kreuzungen monohybrid spalten, war schon MENDEL (1866) bekannt. WHITE (1917) belegte das hierfür verantwortliche Gen mit dem Symbol *Gp*.

Für die Punktierung der Samenschale sind bisher zwei Gene, *F* und *Fs*, bekannt. Beide diese bedingen in dominanter Form anscheinend dieselbe feine, violette Punktierung der Testa. Die Existenz von zwei Genen wurde durch WINGE (1936) nachgewiesen. Er fand in drei Kreuzungen mit zusammen 1145 Individuen Spaltung nach 15 punktiert : 1 nicht punktiert mit D/m = 1,29. Ausserdem wurde von WELLENSIEK (1927, 1929, 1930) und von WINGE (l. c.) Zugehörigkeit von *F* bzw. *Fs* zu verschiedenen Koppelungsgruppen festgestellt.

Die Vererbung der gestreiften Samenschale wurde von LAMPRECHT (1937 a) studiert, der zeigte, dass die Streifung durch Dominanz in einem Gen *Ast* verursacht wird. Die Dominanz scheint vollkommen zu sein.

In bezug auf Koppelung der oben genannten Gene *Cp*, *Gp*, *F*, *Fs* und *Ast* untereinander und mit anderen Genen liegen bisher folgende Feststellungen vor.

Über *das Gen Cp* scheinen Koppelungsstudien nur von WELLENSIEK (1925 a, 1928 a, 1929) vorzuliegen. Er fand freie Kombination von *Cp* zusammen mit den Genen *A*, *M*, *F*, *Oh*, *B*, *P*, *V* und *Pur* (das zweite Gen für Punktierung *Fs* war WELLENSIEK noch unbekannt und *Pur* ging unter der Bezeichnung P_1). Mehr oder weniger deutliche Koppelung gibt WELLENSIEK für *Cp* mit *Gp*, *Bt* und *N* an. Für die Koppelung *Cp*—*Gp* teilt er Spaltungszahlen von 3 Kreuzungen mit zusammen 791 Individuen mit. Die Prozente für das Crossingover betrugen in diesen 34,1, 33,2 und 41,2. Ferner wurden zwei Rückkreuzungen mit zusammen 873 Individuen studiert, die hierfür 38,6 bzw. 40,6 % ergaben. Diese Zahlen scheinen für eine Koppelung zwischen *Cp* und *Gp* zu sprechen; mit Hinblick auf die auch von WELLENSIEK hervorgehobenen Klassifikationsschwierigkeiten bei der Abgrenzung von geraden und gekrümmten Hülsen wird man jedoch unsicher, ob und wie starke Koppelung vorhanden sein kann. — Den von WELLENSIEK für die Koppelung von *Cp* mit *N* mitgeteilten Resultaten dürfte kaum Beweiswert zugesprochen werden können, da — wie bereits E. NILSSON (1939) betont hat — *nCp* und *ncp* Hülsen phänotypisch nicht voneinander unterschieden werden können. — Zwischen Krümmung der Hülse, *Cp*, und *Bt* (*Btb*), stumpfes—spitzes Hülsenende, soll laut WELLENSIEK (l. c.), RASMUSSON (1927) und E. NILSSON (1939) ein deut-

licher Zusammenhang bestehen. Da weiter DE WINTON (laut WINGE, 1936) einen Zusammenhang zwischen *Cp* und *Pa* (hell- und dunkelgrüne Hülsenfarbe) gefunden hat, sollte *Cp* auch mit *Pa* und *Bt* (*Btb*) gekoppelt sein.

Für *das Gen Gp* wurde bisher freie Vererbung im Zusammenhang mit folgenden Genen und von folgenden Verfassern festgestellt. WELLENSIEK (1925 a, 1925 b, 1927, 1928 a, 1929, 1930): *A*, *Ar*, *B*, *M*, *Pl*, *S*, *K*, *I*, *P*, *V*, *N*, *Pur*, *D*, *Fa*, *Tl*, *Le*, *Wb*; NEFF and WHITE (1927): *O*; WINGE (1936): *A*, *Ar*, *B*, *M*, *Pl*, *Z*, *R*, *I*, *P*, *D*, *Fa*, *Le*, *Bl* und *Red*. Für das letztgenannte Gen *Red*, das in rezessiver Form *reduzierte*, schmale Blättchen bedingt, hat WINGE (l. c.) das Symbol *Re* eingeführt. Dieses Symbol (*Re*) kann jedoch nicht beibehalten werden, da es von E. NILSSON (1932) bereits viel früher für ein Gen vergeben worden ist, das in rezessiver Form einen *reproduktionsletalen* Erbsentypus mit herkogamen Blüten bedingt. Ich ändere deshalb WINGEs *Re* zu *Red* ab. Über Koppelungen von *Gp* mit anderen Genen ist ausser der vorhin besprochenen, unsicheren mit *Cp*, noch Koppelung mit einem der Gene für Punktierung der Testa, *F* bzw. *Fs*, bekannt. WELLENSIEK (1928 a) fand hierfür in einer Kreuzung mit 180 F_2-Individuen einen Crossingoverwert von 22,4 %. WINGE (1936), der mit dem Punktierungsgen *Fs* arbeitete, erhielt in einer Kreuzung mit 2441 F_2-Individuen einen Crossingoverwert von 37,4 %, in einer anderen mit 736 Individuen einen solchen von 47,0 %. Der erste Wert dürfte, namentlich mit Hinblick auf die grosse Individuenanzahl und die gleichmässige Verteilung des Ausfalls an Neukombinationen, *Gp Fs* und *gp fs*, zweifellos als sicher zu betrachten sein. Der zweite Wert, 47,0 % besagt dagegen nichts.

Die Gene für Punktierung der Testa, *F* bzw. *Fs*, sind von mehreren Forschern hinsichtlich Koppelung studiert worden. KAJANUS und BERG (1919) und KAJANUS (1923) finden unabhängige Vererbung zusammen mit *A*, *M*, *Oh* und *Pl*; ihre Individuenzahlen sind jedoch zu klein, um einigermassen sichere Schlüsse zu gestatten. H. und O. TEDIN (1928) finden unabhängige Vererbung zusammen mit *A*, *M*, *Fl* und *D*. WELLENSIEK (1927, 1928 a, 1928 b, 1929, 1930) untersuchte die Koppelung zusammen mit folgenden Genen: *B*, *M*, *Pl*, *S*, *K*, *I*, *P*, *V*, *N*, *Cp*, *Gp*, *Pur*, *D*, *Fa*, *Le*, *Tl* und *Wb*. Hierbei fand er in zusammen 6 Kreuzungen mit 4688 Individuen Koppelung für *F*—*B* mit Crossingoverwerten von 8—19 % und für *F*—*M* solche von 33—46 %. *B*—*M* zeigte annähernd 50 % Crossingover. *F* gehört demnach ohne Zweifel zur Koppelungsgruppe *Uni*—*M*—*Mp*—*St*—*Gl*—*B* (vgl. LAMPRECHT, 1933 und 1937 b). In einem anderen, schon oben erwähnten Fall fand

WELLENSIEK Koppelung zwischen dem Punktierungen und der gelben Hülsenfarbe, *Gp*, mit einem Crossingover von 22,4 %. Hier handelte es sich, wie aus dem folgenden hervorgehen wird, wahrscheinlich um das zweite Gen für Punktierung, *Fs*. Für alle übrigen oben aufgezählten Gene fand WELLENSIEK keine Koppelung mit der Punktierung der Testa. — WINGE (l. c.) arbeitete mit dem Punktierungsgen *Fs*. Er fand für dieses, wie bereits mitgeteilt, klare Koppelung mit dem Gen *Gp* für gelbe Hülsenfarbe. Überdies studierte er die Spaltung in *Fs* : *fs* zusammen mit der in folgenden Genen: *Ar*, *B*, *M*, *Pl*, *I*, *R*, *P* und *Red*, die durchweg von *Fs* unabhängige Vererbung zeigten.

Das Gen Ast für Streifung der Testa wurde von LAMPRECHT (1937 a) in bezug auf Koppelung mit den Genen *M*, *Pl*, *V* und *H* studiert. In keinem dieser Fälle konnte sichere Koppelung gefunden werden.

Hinsichtlich Koppelung zwischen den vorstehend besprochenen vier Eigenschaftspaaren, entsprechend den Genen *Cp*, *Gp*, *F*, *Fs* und *Ast*, ist also bisher folgendes bekannt. Eine sichere Koppelung scheint zwischen *Gp* und *Fs* zu bestehen: Crossingover = 37,4 %, festgestellt in einer F_2 von 2441 Individuen. Auch die von WELLENSIEK gefundene Koppelung zwischen *Gp* und Punktierung mit 22,4 % Crossingover dürfte sich auf das Gen *Fs* beziehen. Das zweite Punktierungsgen, *F*, gehört zweifellos einer anderen Koppelungsgruppe an, nämlich *Uni—M—Mp—St—Gl—B*, da zwischen *F* und *B* sichere Koppelung von 8—19 % Crossingover, und zwischen *F* und *M* solche von 33—46 % festgestellt worden ist. Zwischen *Cp* und *Gp* wurde eine Koppelung, entsprechend 34—41 % Crossingover gefunden. Aber wegen der in den betreffenden Kreuzungen aufgetretenen Klassifikationsschwierigkeiten hinsichtlich gerade—gekrümmte Hülse, scheint weder über die Koppelung als solche noch über die Stärke derselben Sicheres ausgesagt werden zu können. WELLENSIEKs Koppelung *Cp—N* kann aus früher angegebenen Gründen nicht als erwiesen betrachtet werden. — In bezug auf die angeführten Koppelungen wäre, wenn es auch unwahrscheinlich sein dürfte, noch an die Möglichkeit des Vorkommens von translozierten Typen mit abweichender Chromosomenstruktur und damit abweichenden Koppelungsverhältnissen zu denken. Angaben über auftretende Semisterilität bzw. Zugehörigkeit der Elternlinien zum Normaltypus fehlen nämlich in den meisten zitierten Arbeiten.

EIGENE UNTERSUCHUNGEN.

Zu Beginn sei hier hervorgehoben, dass in den vier unten zu besprechenden Kreuzungen keine Semisterilität aufgetreten ist und dass sämtliche Elternlinien dem Normaltypus hinsichtlich Chromosomenstruktur angehören.

Kreuzung Nr. 356, ausgeführt zwischen L. 234, *Ast*, und L. 341, *wlo*. Linie 234 hat folgende Eigenschaften, soweit sie hier in Betracht kommen: Samen gelb, nicht runzelig. Testa grünlichgrau, nicht marmoriert, stark gestreift (vgl. LAMPRECHT, 1937 a), schwach blassgraulich oder nicht punktiert (diese Punkte wurden l. c. irrtümlich mit der violetten Punktierung verwechselt und als *F* bezeichnet); Hilum schwarz, Wuchs hoch, ganz wachsig. — L. 341 ist identisch mit der von E. NILSSON (1939) in der Erbsensorte English Wonder entdeckten Pflanze mit lokaler Wachslosigkeit; die Oberseite der Blättchen sind ganz ohne Wachs, während alle übrigen vegetativen Teile stark wachsig sind. Von übrigen Eigenschaften seien erwähnt: Blüten weiss, Kotyledonen grün, Hilum hell, Wuchs niedrig. In Genenformeln ausgedrückt unterscheiden sich die beiden Linien wie folgt:

Linie 234: *A fs Pl Ast I Le Wlo*
» 341: *a Fs pl ast i le wlo*

Die F_1-Generation gab Samen mit Streifung und deutlicher Violettpunktierung. F_2 zeigte folgende Spaltungen:

340 *A*	: 103 *a*	;	D/m	für	3 : 1	= 0,85	
256 *Fs*	: 84 *fs*	;	»	»	»	= 0,12	(nur *A*-Individuen)
327 *Pl*	: 116 *pl*	;	»	»	»	= 0,58	
249 *Ast*	: 91 *ast*	;	»	»	»	= 0,75	(nur *A*-Individuen)
327 *I*	: 116 *i*	;	»	»	»	= 0,58	
332 *Le*	: 111 *le*	;	»	»	»	= 0,03	
342 *Wlo*	: 101 *wlo*	;	»	»	»	= 1,07	

Sämtliche monohybriden Spaltungen zeigten demnach sehr gute Übereinstimmung mit dem theoretisch erwarteten Verhältnis. Von den bifaktoriellen Spaltungen werden nur die mit den hier in Frage stehenden Genen *Fs* und *Ast* mitgeteilt.

Gefunden:	249 *A Ast*	: 91 *A ast*	: 103 *a*	$\left(\frac{Ast}{ast}\right)$
Erwartet:	249,19 »	: 83,06 »	: 110,75 »	
D/m für 9 : 3 : 4 =	— 0,02	+ 0,97	— 0,85	

Gefunden: 256 *A Fs* : 84 *A fs* : 103 $a\left(\frac{Fs}{fs}\right)$
D/m für
9 : 3 : 4 = + 0,65 + 0,11 — 0,85

Gefunden: 167 *Ast Fs* : 82 *Ast fs* : 89 *ast Fs* : 2 *ast f*
Erwartet: 191,25 » : 63,75 » : 63,75 » : 21,25 »
D/m für
9 : 3 : 3 : 1 = — 2,65 + 2,54 + 3,50 — 4,32

Gefunden: 184 *Ast Pl* : 65 *Ast pl* : 64 *ast Pl* : 27 *ast pl*
D/m für
9 : 3 : 3 : 1 = — 0,79 + 0,17 + 0,03 + 1,29

Gefunden: 181 *Ast I* : 68 *Ast i* : 65 *ast I* : 26 *ast i*
D/m für
9 : 3 : 3 : 1 = — 1,12 + 0,59 + 0,17 + 1,06

Gefunden: 193 *Ast Le* : 56 *Ast le* : 62 *ast Le* : 29 *ast le*
D/m für
9 : 3 : 3 : 1 = + 0,19 — 1,08 — 0,24 + 1,74

Gefunden: 194 *Ast Wlo* : 55 *Ast wlo* : 67 *ast Wlo* : 24 *ast wlo*
D/m für
9 : 3 : 3 : 1 = + 0,30 — 1,21 + 0,45 + 0,62

Gefunden: 187 *Fs Pl* : 69 *Fs pl* : 61 *fs Pl* : 24 *fs pl*
D/m für
9 : 3 : 3 : 1 = — 0,46 + 0,73 — 0,38 + 0,39

Gefunden: 185 *Fs I* : 71 *Fs i* : 61 *fs I* : 23 *fs i*
D/m für
9 : 3 : 3 : 1 = — 0,68 + 1,00 — 0,38 + 0,39

Gefunden: 194 *Fs Le* : 62 *Fs le* : 61 *fs Le* : 23 *fs le*
D/m für
9 : 3 : 3 : 1 = + 0,30 — 0,24 — 0,38 + 0,39

Gefunden: 194 *Fs Wlo* : 62 *Fs wlo* : 67 *fs Wlo* : 17 *fs wlo*
D/m für
9 : 3 : 3 : 1 = + 0,30 — 0,24 + 0,45 — 0,95

Die erhaltenen Spaltungszahlen zeigen, dass die beiden Gene *Fs* und *Ast* miteinander stark gekoppelt sind; das Crossingover erreicht einen Wert von etwa 1 %, doch sind diese Werte in Kreuzungen vom Typus $Ab \times aB$ mit recht grossen Fehlern behaftet. Für die bifak-

toriellen Spaltungen der übrigen, zusammen mit *Fs* bzw. *Ast* studierten Gene *A*, *Pl*, *I*, *Le* und *Wlo* wurden Zahlen erhalten, die durchweg für unabhängige Vererbung sprechen.

Da nun WINGE (1936) zwischen dem Punktierungsgen *Fs* und *Gp*, dem Gen für grüne bzw. gelbe Hülsenfarbe, sichere Koppelung mit 37,4 % Crossingover festgestellt hat, war es von Interesse Kreuzungen zu untersuchen, in denen diese beiden Gene spalteten. Da mir nur *a gp*-Linien mit einem Punktierungsgen zur Verfügung standen, von dem unbekannt war ob es *F* oder *Fs* ist, wurde als zweiter Elter eine nicht punktierte, aber marmorierte Linie gewählt, wodurch man auf Grund der Koppelung zwischen *F* und *M* entscheiden können sollte, ob die *a gp*-Linie das Punktierungsgen *F* oder *Fs* enthält.

Kreuzung Nr. 256, ausgeführt zwischen L. 20 und L. 241. Linie 241 stammt aus der deutschen Zuckererbsensorte Goldfähnchen. Sie hat runde, gelbe Samen mit hellem Hilum. Ihre Hülsen sind gelb und haben die Membranformel *pp VV*. Linie 20 stammt aus H. und O. TEDINS (1928) 0651 aus Glaenö. Sie hat runde, gelbe, marmorierte, nicht punktierte Samen mit hellem Hilum; die Samen sind überdies teilfarbig (*z*). Ihre Hülsen sind grün und haben starke Membran, *PP VV*.

Die auf der F_1-Generation erhaltenen Samen waren, wie erwartet, marmoriert und violett punktiert, die Hülsen grün. Die zweite Generation zeigte folgende, hier belangreiche, Spaltungen.

339 *Gp* : 110 *gp*; D/m für 3 : 1 = 0,24
350 *A* : 99 *a* ; » » » = 1,44
355 *M* : 94 *m* ; » » » = 1,99
193 *Fs* : 65 *fs* ; » » » = 0,07 (nur *AZ*-Individuen)
258 *Z* : 92 *z* ; » » » = 0,55 (nur *A*-Individuen)
353 *P* : 96 *p* ; » » » = 1,77

Die gefundenen monohybriden Spaltungsverhältnisse zeigen, wie aus den Werten für D/m hervorgeht, durchweg befriedigende Übereinstimmung mit den theoretisch erwarteten. Für die bifaktoriellen Spaltungen, die hier von Interesse sind, konnten folgende Zahlenverhältnisse festgestellt werden.

Gefunden:	263 *Gp A*	: 76 *Gp a*	: 87 *gp A*	: 23 *gp a*
Erwartet:	252,56 »	: 84,19 »	: 84,19 »	: 28,06 »
D/m für 9 : 3 : 3 : 1 =	+ 0,99	— 0,99	+ 0,34	— 0,99

Gefunden:	273	*Gp M* : 66	*Gp m* : 82	*gp M* : 28	*gp m*
D/m für 9 : 3 : 3 : 1 =	+ 1,95	— 2,20	— 0,26	— 0,01	
Gefunden:	140	*Gp Fs* : 55	*Gp fs* : 53	*gp Fs* : 10	*gp fs* (nur *AZ*-
Erwartet:	145,12	» : 48,37	» : 48,37	» : 16,12	» Indiv.)
D/m für 9 : 3 : 3 : 1 =	— 0,64	+ 1,06	+ 0,74	— 1,57	
Gefunden:	195	*Gp Z* : 68	*Gp z* : 63	*gp Z* : 24	*gp z* (nur *A*-In-
Erwartet:	196,87	» : 65,62	» : 65,62	» : 21,87	» dividuen)
D/m für 9 : 3 : 3 : 1 =	— 0,20	+ 0,33	— 0,36	+ 0,47	
Gefunden:	267	*Gp P* : 72	*Gp p* : 86	*gp P* : 24	*gp p*
D/m für 9 : 3 : 3 : 1 =	+ 1,37	— 1,47	+ 0,22	— 0,79	
Gefunden:	152	*M Fs* : 50	*M fs* : 41	*m Fs* : 15	*m fs* (nur *AZ*-
D/m für 9 : 3 : 3 : 1 =	+ 0,86	+ 0,26	— 1,17	— 0,29	Indiv.)

Von den mitgeteilten bifaktoriellen Spaltungsverhältnissen kann nur eines als Koppelung andeutend aufgefasst werden; es betrifft dies die schon von WINGE festgestellte Koppelung *Gp—Fs*. Den hierfür erhaltenen Zahlen entspricht ein Crossingover von 40,5 %, während WINGE 37,4 % gefunden hat. WINGEs Zahlen sind gross genug (2441 Individuen) um die Koppelung sicher zu beweisen, meine obigen können sie nur als wahrscheinlich erscheinen lassen. Dass es sich in meiner Kreuzung um das Punktierungsgen *Fs*, wie bei WINGE, und nicht um *F* handelt, dafür spricht, dass zwischen *M* und *Fs* keine Andeutung von Koppelung gefunden worden ist, während *F* mit *M* Crossingoverwerte von etwa 39 % gibt.

Kreuzung Nr. 361, ausgeführt zwischen L. 241 und L. 456. Diese Kreuzung wird hier mitgeteilt um eine weitere Bestätigung der in voriger Kr. 256 gefundenen Koppelung zwischen *Gp* und *Fs* zu erhalten. Linie 241 wurde schon oben beschrieben. Line 456 stammt aus meiner Kreuzung Nr. 25, ausgeführt zwischen L. 5, Chenille und L. 102, Acacia. Es kommt ihr folgende Genenformel zu: *A m fs Z oh pl s ast r i le tl wb*. Unten werden nur jene Spaltungsresultate mitgeteilt, die für die vorliegende Arbeit von Interesse sind. Die auf F_1 erhaltenen Samen waren rund und hatten, wie erwartet, violettpunktierte Testa

sowie helles Hilum. Die Hülsen der F_1 waren grün. In F_2 wurden für die Gene *Gp*, *A* und *Fs* folgende mono- und bifaktorielle Spaltungen festgestellt.

293 *Gp* : 92 *gp*; D/m für 3 : 1 = 0,50
285 *A* : 100 *a* ; » » » = 0,44
209 *Fs* : 76 *fs* ; » » » = 0,65 (nur *A*-Individuen).

Bifaktorielle Spaltungen:

Gefunden:	213 *Gp A*	: 80 *Gp a*	: 72 *gp A*	: 20 *gp a*	
Erwartet:	216,00 »	: 72,19 »	: 72,19 »	: 24,06 »	
D/m für 9 : 3 : 3 : 1 =	— 0,36	+ 1,02	— 0,02	— 0,85	
Gefunden:	151 *Gp Fs*	: 62 *Gp fs*	: 58 *gp Fs*	: 14 *gp fs*	(nur *A*-
Erwartet:	160,31 »	: 53,44 »	: 53,44 »	: 17,81 »	Indiv.)
D/m für 9 : 3 : 3 : 1 =	— 1,11	+ 1,30	+ 0,69	— 0,93	

Für die Koppelung zwischen den beiden Genen *Gp* und *Fs* ergibt sich in dieser Kreuzung praktisch genommen ganz derselbe Wert wie in der vorigen, nämlich ein Crossingover von 39,7 % gegenüber 40,5 % in Kr. 256. Da beide Kreuzungen den einen Elter, L. 241, mit den Genen *gp* und *Fs* gemeinsam haben, können die für diese Gene gefundenen Spaltungsverhältnisse vereinigt werden. Es ergibt sich hierbei:

Gefunden:	291 *Gp Fs*	: 117 *Gp fs*	: 111 *gp Fs*	: 24 *gp fs*	
Erwartet:	305,44 »	: 101,81 »	: 101,81 »	: 33,94 »	
D/m für 9 : 3 : 3 : 1 =	— 1,25	+ 1,67	+ 1,12	— 1,76	

Aus diesen Zahlen berechnet sich ein Crossingoverwert von 40,0 %, der mit WINGES Wert von 37,4 % gute Übereinstimmung zeigt.

Da nun sowohl *Gp* mit *Fs* wie *Fs* mit *Ast* als sicher gekoppelt aufgefasst werden konnten, war die Untersuchung einer Kreuzung wünschenswert, die teils in diesen drei Genen, teils wenn möglich auch in *Cp* spaltete, da eine Koppelung von *Gp* mit *Cp* von WELLENSIEK in Frage gestellt worden ist. Dieser Forderung entspricht:

Kreuzung Nr. 272, ausgeführt zwischen L. 234 und L. 241, die beide schon bei der Besprechung vorstehender Kreuzungen charakterisiert worden sind. Hinzuzufügen ist hier nur, dass L. 241 deutlich gekrümmte (*cp*), L. 234 gerade Hülsen hat (*Cp*). Hier hat nur die

Spaltung in folgenden Genen Interesse: *Cp*, *Gp*, *A*, *Fs* und *Ast*. Die Spaltung in *A* wird, wie schon oben erwähnt, angeführt, um Koppelung zwischen diesem Gen und *Fs* bzw. *Ast* ausschliessen zu können, da letztere ja nur an *A*-Individuen beurteilt werden können. Die auf F_1 geernteten Samen zeigten, wie erwartet, sowohl gestreifte wie violett punktierte Testa. Die Hülsen der F_1-Pflanzen waren gerade und grün.

Die zweite Generation zeigte in den genannten fünf Genen folgende Spaltungen:

305 *Cp* : 107 *cp* ; D/m für 3 : 1 = 0,45
312 *Gp* : 100 *gp* ; » » » = 0,34
307 *A* : 105 *a* ; » » » = 0,23
232 *Fs* : 75 *fs* ; » » » = 0,23 (nur *A*-Individuen)
228 *Ast* : 79 *ast*; » » » = 0,29 (nur *A*-Individuen)

In bezug auf die Beurteilung des Eigenschaftspaares gerade—gekrümmte Hülse ist hier besonders hervorzuheben, dass diese in vorliegender Kreuzung keinerlei Schwierigkeit verursachte; nur zwei Individuen hatten Hülsen, die als *cp*(?) registriert wurden. Es wurde demnach für das Genpaar *Cp*—*cp* ein gutes monohybrides Spaltungsverhältnis gefunden; D/m = 0,45. Für die hier interessierenden Gene wurden folgende bifaktorielle Spaltungen in F_2, F_3 und F_4 festgestellt.

F_2, gefunden:	302	*Cp Gp*:	2	*Cp gp* :	10	*cp Gp*:	73	*cp gp*
F_3+F_4 » :	303	» :	10	» :	3	» :	92	»
Zusammen :	605	*Cp Gp*:	12	*Cp gp*:	13	*cp Gp*:	165	*cp gp*
Erwartet :	447,19	» :	149,06	» :	149,06	» :	49,69	»
D/m für 9:3:3:1 =	+11,3		− 12,5		− 12,4		+ 16,9	

F_2, gefunden:	159	*Cp Fs* :	65	*Cp fs* :	73	*cp Fs*:	10	*cp fs*
F_3+F_4 » :	83	» :	38	» :	33	» :	5	»
Zusammen :	242	*Cp Fs* :	103	*Cp fs* :	106	*cp Fs*:	15	*cp fs*
Erwartet :	262,12	» :	87,37	» :	87,37	» :	29,12	»
D/m für 9:3:3:1 =	− 1,88		+ 1,86		+ 2,21		− 2,71	

F_2, gefunden:	178	*Cp Ast*:	46	*Cp ast*:	49	*cp Ast*:	34	*cp ast*
F_3+F_4 » :	38	» :	30	» :	11	» :	20	»
Zusammen :	216	*Cp Ast*:	76	*Cp ast*:	60	*cp Ast*:	54	*cp ast*

Erwartet	:	228,37	*Cp Ast*:	76,12	*Cp ast*:	76,12	*cp Ast*:	25,37	*cp ast*
D/m für										
9:3:3:1 =		— 1,24		— 0,02		— 2,05		+ 5,88		
F_2, gefunden:		163	*Gp Fs*:	70	*Gp fs*:	69	*gp Fs*:	5	*gp fs*
F_3+F_4 »	:	72	» :	28	» :	28	» :	3	»
Zusammen	:	235	*Gp Fs*:	98	*Gp fs*:	97	*gp Fs*:	8	*gp fs*
Erwartet	:	246,37	» :	82,12	» :	82,12	» :	27,37	»
D/m für										
9:3:3:1 =		— 1,10		+ 1,94		+ 1,82		— 3,82		
F_2, gefunden:		184	*Gp Ast*:	49	*Gp ast*:	43	*gp Ast*:	31	*gp ast*
F_3+F_4 »	:	45	» :	22	» :	7	» :	17	»
Zusammen	:	229	*Gp Ast*:	71	*Gp ast*:	50	*gp Ast*:	48	*gp ast*
Erwartet	:	223,87	» :	74,62	» :	74,62	» :	24,87	»
D/m für										
9:3:3:1 =		+ 0,52		— 0,48		— 3,17		+ 4,79		
F_2, gefunden:		152	*Fs Ast*:	80	*Fs ast*:	75	*fs Ast*:	0	*fs ast*
F_3+F_4 »	:	148	» :	145	» :	118	» :	1	»
Zusammen	:	300	*Fs Ast*:	225	*Fs ast*:	193	*fs Ast*:	1	*fs ast*
Erwartet	:	404,44	» :	134,81	» :	134,81	» :	44,94	»
D/m für										
9:3:3:1 =		— 7,85		+ 8,62		+ 5,56		— 6,77		

Die in F_2 und $F_3 + F_4$ erhaltenen Spaltungen zeigen meistens gute, stets aber befriedigende Übereinstimmung. Sie zeigen ganz eindeutig, dass die hier studierten vier Gene, *Cp*, *Gp*, *Fs* und *Ast*, zu einer Koppelungsgruppe gehören. Eine Berechnung der Crossingoverwerte ergibt folgendes:

Cp—Gp = 3,2 %	Crossingover	(Individuenanzahl	795)
Cp—Fs = 32,2 %	»	(»	466)
Cp—Ast = 42,5 %	»	(»	406)
Gp—Fs = 36,0 %	»	(»	438)
Gp—Ast = 37,4 %	»	(»	398)
Fs—Ast = <1 %	»	(»	719)

SCHLUSSBETRACHTUNG.

Eine Zusammenfassung der Ergebnisse der vorstehend besprochenen vier Kreuzungen ergibt folgendes. Die Gene *Gp*, *Cp*, *Fs* und *Ast* bilden eine Koppelungsgruppe. Die Koppelung zwischen *Fs* und *Ast*

ist am stärksten. Sie wurde in zwei Kreuzungen mit zusammen 1321 Individuen studiert und entsprach einem Crossingover von weniger als 1 %. Die zunächst stärkste Koppelung zeigten die Gene *Cp* und *Gp* mit 3,2 % Crossingover. Die umfangreichsten Untersuchungen liegen für die Koppelungsgruppe *Gp—Fs* vor. In der oben besprochenen Kreuzung Nr. 272 mit 981 Individuen resultierte ein Crossingover von 37,1 %. WINGE (1936) hat in einer Kreuzung mit 2441 Individuen 37,4 % gefunden. Für die Koppelung zwischen *Cp* und *Fs* ergab sich ein Crossingoverwert von 32,2 %, für die zwischen *Gp* und *Ast* ein solcher von 37,4 % und für die zwischen *Cp* und *Ast* ein solcher von 42,5 %. Auf Grund dieser Zahlen erscheint die in Fig. 1 wiedergegebene Verteilung der vier Gene *Gp—Cp—Fs—Ast* im Chromosom als die wahrscheinlichste. Als vollkommen sicher ist sie jedoch nicht zu betrachten, da die beiden Gene *Gp* und *Cp* einerseits und besonders *Fs*

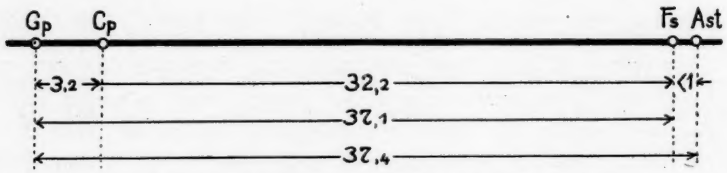

Fig. 1. Die Koppelungsgruppe *Gp—Cp—Fs—Ast*.

und *Ast* andererseits sehr nahe einander gelegen sind. Für diese beiden Genpaare kann daher eine umgekehrte Anordnung auf Grund des bisher vorliegenden Zahlenmaterials noch nicht ausgeschlossen werden.

E. NILSSON (1939) hat auf Grund der Resultate von WELLENSIEK, RASMUSSON und DE WINTON, wie früher erwähnt, die Zugehörigkeit von *Cp* (bzw. eines Gens für Krümmung der Hülse) zur Koppelungsgruppe *Pa—R—Tl—Bt* vermutet. Die oben besprochene Kreuzung Nr. 356 spaltete auch in *R*, doch zeigte dieses Gen keine sichere Koppelung mit *Fs* und Kreuzung Nr. 361 spaltete in den beiden stark gekoppelten Genen *R* und *Tl*, ohne dass diese eine Andeutung zu Koppelung mit *Gp*, mit welchem Gen *Cp* stark gekoppelt ist, zeigten. Die Zugehörigkeit der Koppelungsgruppe *Gp—Cp—Fs—Ast* zu *Pa—R—Tl—Bt* dürfte daher wohl bis auf weiteres als ungeklärt zu betrachten sein. Die Erklärung der von NILSSON angeführten Beobachtungen könnte vielleicht in einem zweiten Gen für Krümmung der Hülse zu suchen sein.

SUMMARY.

1. The four pairs of properties: straight—curved pod, green—yellow pod-colour, violet—non-speckling, and striation—non-striation of the seed-coat were studied as to their inheritance.

2. It is shown that the four genes *Cp*, *Gp*, *Fs* and *Ast*, which cause these properties, belong to the same linkage-group. The degree of linkage between these genes is illustrated by Fig. 1.

3. For the speckling of the seed-coat there exist two genes with the same effect, *F* and *Fs*. The gene *F* belongs to the linkage-group *Uni—M—Mp—St—Gl—B*.

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GENSTUDIEN AN PISUM SATIVUM

V. MULTIPLE ALLELE FÜR PUNKTIERUNG DER TESTA: Fs_{ex}—Fs—fs

VON *HERBERT LAMPRECHT*

SAATZUCHTANSTALT WEIBULLSHOLM, LANDSKRONA

(With a Summary in English)

EINLEITUNG.

FÜR die Punktierung der Samenschale von *Pisum sativum* werden gegenwärtig zwei Gene, *F* und *Fs*, verantwortlich gemacht. Beide diese Gene scheinen in dominanter Form ganz den gleichen Typus von feiner, violetter Punktierung zu verursachen. Sowohl *F* wie *Fs* wirken nur bei Dominanz im Grundgen für Farbenausbildung *A*.

LOCK (1904) fand zuerst in einer Kreuzung Spaltung nach 3 punktiert : 1 nicht punktiert. E. v. TSCHERMAK (1912) konnte dies bestätigen. Letzterer verwendete für das hierfür verantwortliche Gen das Symbol *F*. Ausserdem machte er die Wirkung von *F* noch von einem weiteren Gen *E* abhängig, das mit *A* absolut gekoppelt sein sollte. Dieses hypothetische Gen *E* wurde später, als zum pleiotropen Wirkungsbereich von *A* gehörig, gestrichen (vgl. WELLENSIEK, 1925).

WELLENSIEK (1927, 1929, 1930) stellte als erster sichere Koppelung von *F* mit *B* und *M* fest. *B* bedingt zusammen mit *A* und *Ar* purpurfarbige Blüte und *M* verursacht sowohl bei *A* wie *a* Marmorierung der Testa. Für die Koppelung *F*—*B* wurden Crossingoverwerte von 8—17 % erhalten, für die Koppelung *F*—*M* solche von 33—46 %. Dies wurde von WELLENSIEK in 6 Kreuzungen mit zusammen 4688 Individuen klar festgestellt. Danach kann kein Zweifel bestehen, dass das von diesem Forscher studierte Punktierungsgen *F* zur Koppelungsgruppe *Uni—M—Mp—St—Gl—B*, also zum *B*-Chromosom von *Pisum*, wie ich es nennen will, gehört. Hier sei — ohne die durch WELLENSIEK festgestellte Koppelung irgendwie bezweifeln zu wollen — hervorgehoben, dass ich in verschiedenem Kreuzungs- und Linienmaterial die Beobachtung gemacht habe, dass die Punktierung der Testa bei *A Ar b*-Samen, also bei Pflanzen mit rosa Blütenfarbe, häufig sehr schwach oder gar nicht erkennbar ist, wodurch für solche Pflanzen ein deutliches Defizit an *F*-Pflanzen entstehen kann. Vielleicht ist dies von der

übrigen genotypischen Konstitution in nicht geringem Grade abhängig. Auch ist noch nicht klargelegt, ob sich diese Erscheinung auf *F* oder *Fs* bezieht.

WINGE (1936) fand in vier Kreuzungen mit zusammen 2441 Individuen Koppelung zwischen den Eigenschaften punktierte Testa und gelbe Hülsenfarbe. Letztere wird durch das rezessive Gen *gp* bedingt (s. WHITE, 1917). Als Crossingoverwerte wurden in diesen Kreuzungen, die alle der Kombination $Ab \times aB$ entsprachen, Werte von 24,5 bis 43,5 % oder durchschnittlich 37,4 % gefunden. In einer anderen Kreuzung, die der Kombination $AB \times ab$ entsprach, betrug der entsprechende Wert 47,0 %; dieser kann wegen seiner grossen Nähe zu 50 % nicht als für Koppelung sprechend aufgefasst werden. WINGE hat aber überdies in drei Kreuzungen mit zusammen 1145 Individuen Spaltung nach 1084 punktiert : 61 nicht punktiert, also sehr gut mit 15 : 1 übereinstimmend ($D/m = 1{,}29$) gefunden (l. c. S. 284). Mit Hinblick auf diese Verhältnisse nimmt WINGE an, dass es ausser *F* noch ein zweites Gen für die Ausbildung von Punktierung geben muss. Er belegte dieses mit dem Symbol *Fs*. Die Aufstellung dieses Gens mit Hinblick auf die abweichende Koppelung allein könnte allerdings nicht als einwandfrei anerkannt werden, da es sich ja bei WINGES Linien um einen vom Normaltypus abweichenden translozierten Typus mit anderer Chromosomenstruktur handeln könnte. Und Angaben hierüber bzw. über ev. aufgetretene Semisterilität finden sich in WINGES Arbeit nicht.

EIGENE UNTERSUCHUNGEN.

Die unten zu besprechenden Kreuzungsstudien wurden begonnen, um die Genetik eines Erbsentypus klarzulegen, bei dem die violetten Punkte auf der Testa aussergewöhnlich gross waren. Fig. 1 zeigt den Unterschied zwischen dem gewöhnlichen Typus, links, und dem mit zu kleinen Fleckchen erweiterten Punkten, rechts. Der letztere Typus entstammt einer im nördlichen Schweden, in der Provinz Jämtland, unter dem Namen Grauerbse gebauten Ackererbse. Sie wird hier als Linie 378 geführt. Mit Hinblick auf das bisher über die Vererbung der Punktierung Bekannte wurde mit der Möglichkeit gerechnet, teils dass die beiden Gene *F* und *Fs* zusammen die stärkere Punktierung bedingen, teils dass es ein Modifikationsgen gibt, dass die Vergrösserung der Punkte verursacht. Es wurden mehrere Kreuzungen zwischen sowohl normal punktierten wie nicht punktierten und Linie 378 ausge-

führt. Im folgenden werden vier derselben besprochen, die teils zur Feststellung von drei multiplen Allelen für die Punktierung der Testa, teils zur Klarlegung, ob es sich um das Gen *F* oder *Fs* handelt, geführt haben. Sämtliche zu diesen Kreuzungen verwendeten Linien, L. 19, L. 232, L. 234 und L. 378 scheinen hinsichtlich Chromosomenstruktur dem Normaltypus anzugehören, da sie in allen Kreuzungen mit diesem wie auch untereinander niemals Anzeichen für Semi- oder andere partielle Sterilität gezeigt haben.

Kreuzung Nr. 324, ausgeführt zwischen L. 378 und L. 19. L. 378 ist die vorhin erwähnte Grauerbse (Fig. 1 rechts) mit den starken Punkten auf der Testa; sie hat überdies noch schwarzes Hilum. L. 19 ist identisch mit H. und O. TEDINS (1928) 0652 und stammt aus der

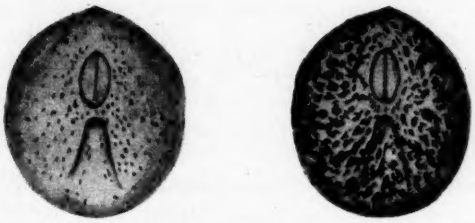

Fig. 1. Links normale Violettpunktierung der Testa, verursacht durch das Gen *Fs* bzw. *F*; rechts starke Violettpunktierung, verursacht durch das dominante Allel Fs_{ex}.

dänischen Ackererbse Glaenö. L. 19 hat zum Unterschied von L. 378 helles Hilum, marmorierte, aber nicht punktierte Samenschale.

Die auf F_1 geernteten Samen hatten, wie erwartet, schwarzes Hilum und waren marmoriert sowie stark punktiert. Die Punktierung war ausgesprochen stark, bedeutend stärker als die durch *F* bzw. *Fs* bedingte, aber nicht ganz so stark wie die des einen Elters, L. 378. Die starke Punktierung dominiert also deutlich, aber nicht vollkommen.

In der zweiten Generation wurden u. a. folgende Spaltungen beobachtet:

Gefunden: 241 *M* (marmoriert) : 74 *m* (nicht marmoriert)
Erwartet: 236,25 » » : 78,75 » » »
D/m für 3 : 1 = 0,62.

Gefunden: 230 *Pl* (schwarzes Hilum) : 85 *pl* (helles Hilum)
D/m für 3 : 1 = 0,81.

Gefunden: 224 stark punktiert : 91 nicht punktiert
D/m für 3 : 1 = 1,60.

Das letzte Spaltungsverhältnis zeigt also, dass die starke Punktierung der Testa von L. 378 nicht auf das Zusammenwirken von zwei Genen zurückzuführen ist, die etwa je für sich schwache Punktierung bedingen, so wie als Arbeitshypothese oben angenommen worden ist. Es besteht nun noch die Möglichkeit, dass die starke Punktierung auf die Wirkung eines Gens für Verstärkung der Punkte zurückzuführen ist, das solchenfalls allein keine Wirkung auf die Testa hätte und auch im anderen Elter, L. 19, in dominanter Form vorhanden sein müsste.

Eine Unterscheidung der stark punktierten Homo- und Heterozygoten, von denen wie erwähnt letztere nicht so starke Punkte aufweisen (F_1), war in F_2 nicht sicher möglich; sie mussten daher bei der Klassifikation vereint werden. Für bifaktorielle Spaltungen wurden folgende Zahlenverhältnisse erhalten.

Gefunden:	174 *M Pl*	: 67 *M pl*	: 56 *m Pl*	: 18 *m pl*
Erwartet:	177,19 »	: 59,06 »	: 59,06 »	: 19,69 »
D/m für 9 : 3 : 3 : 1 =	— 0,36	+ 1,15	— 0,44	— 0,39

Gefunden: 174 *M* punkt. : 67 *M* nicht punkt. : 50 *m* punkt. : 24 *m* nicht punkt.

D/m für 9 : 3 : 3 : 1 = — 0,36, + 1,15, — 1,31, + 1,00

Gefunden: 165 *Pl* punkt. : 65 *Pl* nicht punkt. : 59 *pl* punkt. : 26 *pl* nicht punkt.

D/m für 9 : 3 : 3 : 1 = — 1,38, + 0,86, — 0,01, + 1,47.

Wie ersichtlich spricht keines dieser drei Spaltungsverhältnisse für Koppelung. Da das eine bekannte Gen für Punktierung der Testa, *F*, zur gleichen Koppelungsgruppe wie *M* gehört (s. oben) und die beiden Eltern bei Kreuzung mit dem *Pisum*-Typus von normaler Chromosomenstruktur keine Semisterilität zeigten, erscheint es sehr wahrscheinlich, dass die starke Testapunktierung von L. 378 nicht durch das Gen *F* und ein weiteres Verstärkungsgen bedingt wird. Die nächste Kreuzung wird diesbezüglich Klarheit bringen.

Kreuzung Nr. 357, ausgeführt zwischen L. 378, Grauerbse, und L. 234, Gestreifte, nicht punktierte Testa. L. 234 ist dominant im Gen *Ast* für gestreifte Testa (s. LAMPRECHT, 1937) und rezessiv im Gen *D*, das in dominanter Form die Ausbildung des anthozyanfarbigen Ringes um die Stipelbasis bedingt.

Die auf F_1 geernteten Samen waren, wie erwartet, sowohl gestreift

wie stark punktiert. Die zweite Generation gab für Punktierung und Streifung folgende Spaltungszahlen.

Gefunden: 324 stark punktiert : 112 nicht punktiert
Erwartet: 327,0 » » : 109,0 » »
D/m für 3 : 1 = 0,33.
Gefunden: 353 *Ast* : 83 *ast*
D/m für 3 : 1 = 2,38.

Das für die starke Punktierung gefundene monohybride Spaltungsverhältnis zeigt in Übereinstimmung mit der vorigen Kreuzung, dass es sich nur um Spaltung in einem Gen handeln kann. I. ü. gilt das bereits dort Angeführte. Die Spaltung im Genpaar *Ast—ast* zeigt weniger gute, aber noch befriedigende Übereinstimmung mit dem monohybriden, erwarteten Spaltungsverhältnis. Es handelte sich wahrscheinlich um einen geringeren Ausfall von — vielleicht weniger vitalen — Rezessiven. Für die Spaltung in beiden Genpaaren, *Ast—ast* und dem für die starke Punktierung, ergab sich folgendes Verhältnis.

Gefunden: 241 *Ast* stark punkt. : 112 *Ast* nicht punkt. : 83 *ast* stark punkt. : 0 *ast* nicht punkt.
Erwartet: 245,25 *Ast* stark punkt. : 81,75 *Ast* nicht punkt. : 81,75 *ast* stark punkt. : 27,25 *ast* nicht punkt.
D/m für 9 : 3 : 3 : 1 = — 0,32, + 3,99, + 0,13, — 5,40.

Dieses Verhältnis zeigt zweifellos, dass zwischen den beiden hier in Rede stehenden Genen sehr starke Koppelung besteht. Sie dürfte einem Crossingover von weniger als 5 % entsprechen. In bezug auf die Stärke dieser Koppelung sei hier auf eine andere, gleichzeitig mit dieser erscheinende Arbeit (LAMPRECHT, 1942) verwiesen, in der die Resultate weiterer Kreuzungen, mit Spaltung in den beiden in Rede stehenden Genpaaren, behandelt werden. Da dort auch nachgewiesen wird, dass das Gen *Ast* der l. c. festgestellten Koppelungsgruppe *Gp—Cp—Fs—Ast* angehört, in der *Fs* mit *Ast* etwa dieselbe starke Koppelung mit *Ast* wie in der eben besprochenen Kreuzung Nr. 357 zeigt, kann darauf geschlossen werden, dass das hier in Frage stehende Gen für *starke* Punktierung der Testa höchst wahrscheinlich ein multiples Allel von *Fs* darstellt. Die beiden nun zu besprechenden Kreuzungen werden den exakten Beweis hierfür erbringen.

Kreuzung Nr. 352, ausgeführt zwischen L. 378 aus Grauerbse und L. 232. Letztere Linie stammt von der englischen Erblichkeitsforscherin

DE WINTON. Sie ist u. a. in den Genen *b*, *m*, *mp*, *st*, *k*, *s*, *pl* und *wb* rezessiv. Die Testa zeigt sehr schwach ausgebildete feine Violettpunktierung, die nicht selten kaum zu erkennen ist oder ganz zu fehlen scheint. In einer früheren Arbeit (LAMPRECHT und ÅKERBERG, 1939, S. 333) wurde sie daher irrtümlich als *f*, also mit nicht punktierter Testa klassifiziert. Es stellte sich dann heraus, dass diese schlechte Ausbildung, bzw. das gänzliche Fehlen der Punktierung mit der Rezessivität im Gen *b* zusammenhängt. Linien mit rosa Blütenfarbe, also *A Ar b*, zeigen in der Regel diese Erscheinung mehr oder weniger deutlich.

Die auf F_1 von Kreuzung Nr. 352 erhaltenen Samen zeigten wiederum wie erwartet stark punktierte Samenschale und schwarzes Hilum. Die zweite Generation spaltete hinsichtlich Punktierung wie folgt.

Gefunden: 332 stark punktiert : 109 schwach punktiert
Erwartet: 330,75 » » : 110,25 » »
D/m für 3 : 1 = 0,12.

Während in den beiden früheren Kreuzungen Nr. 324 und 357 monohybride Spaltung nach 3 stark punktiert : 1 nicht punktiert gefunden wurde, zeigt die vorliegende Kreuzung Spaltung nach 3 stark punktiert : 1 schwach punktiert. Das für diese Spaltung verantwortliche Gen zeigte mit keinem der oben für L. 232 erwähnten Gene sichere Koppelung, weshalb das die schwache Punktierung bedingende Gen nicht mit *F*, das zur Koppelungsgruppe *Uni—M—Mp—St—Gl—B* gehört, identisch sein kann. Die Ergebnisse von Kr. 352 machen es also wahrscheinlich, dass das Punktierungsgen von L. 232 mit *Fs* identisch ist. Mit Hinblick auf die in dieser und den vorherigen Kreuzungen erhaltenen Ergebnisse wäre demnach zu erwarten, dass das Gen für die starke Punktierung der Testa von L. 378, Grauerbse, ein weiteres, dominantes Allel des Gens *Fs* darstellen sollte. Das fehlende Glied in der Beweiskette hierfür wird die Kreuzung zwischen den Linien 19 und 232, dem einen Elter der oben besprochenen Kreuzungen Nr. 324 bzw. 352 liefern. Es ist dies

Kreuzung Nr. 246. In bezug auf Beschreibung der Elternlinien siehe die früheren Kreuzungen. Die auf F_1 dieser Kreuzung erhaltenen Samen zeigten die schwache Violettpunktierung. Schon dadurch ist bewiesen, dass der Elter mit nicht punktierter Testa, L. 19, nicht Träger eines Verstärkungsgens für die Punktierung sein kann, wie in Kr. 324 als eventuelle Erklärungsmöglichkeit für die monohybride Spaltung nach 3 stark punktiert : 1 nicht punktiert hervorgehoben worden ist.

In F_2 resultierte folgende Spaltung:

Gefunden: 206 schwach punktiert : 58 nicht punktiert
Erwartet: 198,0 » » : 66,0 » »
D/m für 3 : 1 = 1,13.

In bezug auf die in dieser Kreuzung beobachteten Spaltungsresultate sei noch erwähnt, dass die drei spaltenden Gene *B*, *M* und *St*, die sämtliche zur selben Koppelungsgruppe, nämlich *Uni—M—Mp—St—Gl—B*, gehören, keine Koppelung mit dem Gen gezeigt haben, das hier für die Punktierung verantwortlich ist. Es kann sich demnach nicht um das Gen *F* gehandelt haben, das auch zu dieser Koppelungsgruppe gehört.

SCHLUSSBETRACHTUNG.

Von den mitgeteilten vier Kreuzungen sind drei zwischen den Linien 19, nicht punktiert, 232, schwach punktiert, und 378, stark punktiert, in den drei möglichen Kombinationen ausgeführt worden. Da alle diese drei Kreuzungen monohybride Spaltung nach 3 : 1 zeigten, ist bewiesen, dass die drei Eigenschaften nicht punktierte, punktierte und stark punktierte Samenschale durch diesen entsprechende, multiple Allele bedingt werden. Die vierte Kreuzung, Nr. 357, sowie in einer anderen Arbeit veröffentlichte (LAMPRECHT, 1942) zeigten, dass es sich hierbei um Allele des Gens *Fs* handelt, das in diesen starke Koppelung mit dem Gen *Ast* der Koppelungsgruppe *Gp—Cp—Fs—Ast* zeigte. Das in vorliegender Arbeit neu festgestellte, über *Fs* dominierende Allel, das die starke Punktierung der Linie 378, Grauerbse, bedingt, belege ich mit dem Symbol Fs_{ex}. Das Suffix *ex* wird hierbei, abgeleitet von *expandere* = ausbreiten, mit Hinblick auf die Vergrösserung der Punkte gewählt.

SUMMARY.

1. A new type of speckling of the seed-coat with specially large spots is described (see Fig. 1) and its inheritance investigated.

2. It has been ascertained that the large-spotted seed-coat is caused by a new, hitherto unknown allel of the gene-pair *Fs—fs*. This is designated by Fs_{ex}; the index *ex* is derived from *expandere* = to enlarge. Thus a new series of multiple allels in *Pisum* has been found: Fs_{ex}—*Fs—fs*.

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THE RELATION OF MITOTIC DISTURBANCES TO X-RAY DOSAGE AND POLYPLOIDY

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THE amount of mitotic disturbances, induced in dormant seeds by different X-ray dosages, was recently examined in five species of wheat and oats, namely *Triticum monococcum*, *T. dicoccum*, *T. durum*, *T. vulgare* and *Avena sativa* (FRÖIER, GELIN and GUSTAFSSON, 1941), the first species being diploid, the two following tetraploid and the two last-mentioned hexaploid. The result of this cytological analysis is presented in Table 1.

The data strongly indicate two facts: 1) The percentage of disturbed cell divisions does not increase in direct proportion to the increase in dosage. 2) The rate of increase in percentage of disturbed divisions is not the same in diploids, tetraploids and hexaploids. In the paper mentioned no test was made of the statistical significance of the differences observed. As such a test was considered desirable, in view of the interest at present devoted to the problems concerning X-ray influence upon cell division and mutation, one of the present authors (TEDIN) was invited to join in the collaboration.

The statistical methods presented below are not new. As far as we know, they have not, however, been applied to material of the present type, and as they seem to give some promise of usefulness in such material, we have considered it justified to present the analysis in detail.

The material under discussion affords a case of alternate variation between undisturbed and disturbed cells. The obvious statistical method of analysis must, therefore, be an application of the χ^2-test to the differences observed. This application, however, presents certain difficulties. For each species there are three pairs of values available (disregarding the dosages of 20000 and 25000 r), and it is easy to prove, for each species, that the proportions of disturbed cells are different at different dosages. We have not been able, however, to find any method by which it was possible to test, from the data presented, whether the changes in proportion to increased dosage were the same

TABLE 1. *The number of induced mitotic disturbances in relation to polyploidy and dosage.*

Species	Dose	Number of roots examined	Percentage of disturbed cells
Triticum monococcum.....	5000 r	10	$\frac{284}{2586} = 10{,}98$ %
	10000 r	10	$\frac{498}{1630} = 30{,}55$ %
	15000 r	10	$\frac{1004}{1466} = 68{,}49$ %
Triticum dicoccum	5000 r	10	$\frac{384}{1573} = 24{,}41$ %
	10000 r	8	$\frac{968}{1357} = 71{,}33$ %
	15000 r	7	$\frac{1091}{1257} = 86{,}79$ %
Triticum durum.....	5000 r	11	$\frac{386}{1932} = 19{,}98$ %
	10000 r	6	$\frac{757}{1306} = 57{,}89$ %
	15000 r	7	$\frac{1038}{1248} = 83{,}17$ %
	20000 r	7	$\frac{1005}{1063} = 94{,}54$ %
	25000 r	10	$\frac{1088}{1102} = 98{,}73$ %
Triticum vulgare	5000 r	10	$\frac{422}{1438} = 29{,}35$ %
	10000 r	10	$\frac{810}{1313} = 61{,}69$ %
	15000 r	10	$\frac{1241}{1519} = 81{,}70$ %
	20000 r	4	$\frac{462}{492} = 93{,}90$ %
	25000 r	3	$\frac{136}{142} = 95{,}77$ %
Avena sativa	5000 r	14	$\frac{302}{1451} = 20{,}81$ %
	10000 r	11	$\frac{713}{1679} = 42{,}47$ %
	15000 r	11	$\frac{945}{1350} = 70{,}00$ %
	20000 r	13	$\frac{937}{1035} = 90{,}53$ %
	25000 r	9	$\frac{983}{1024} = 96{,}00$ %

in all species, nor a method to test whether, within each species, the proportions were in agreement with any given hypothesis, such as that of a straight-line proportionality.

If the pair of data for each species and dosage could be reduced to one single value, an adequate χ^2-test could be made. Such a reduction may be obtained by calculating, for instance, percentages of disturbed cells, but the χ^2-test is applicable only to absolute values. The single possible way seemed to be the following: *if exactly the same number of cells is studied in each species-dosage combination*, the relative rates of disturbances may be correctly expressed simply by the number of disturbed cells observed. It should be pointed out that, from the statistical point of view, such a limitation of the material is essential. When, for instance, 2586 cells were classified in *Triticum monococcum*, the seeds having obtained 5000 r, but only 1248 cells in *Triticum durum* after 15000 r, then, from the present viewpoint, the classification of 1338 cells in the former material was made in vain.

In the present material it was possible, by using the original notations, to extract the number of disturbed divisions found among the 1200 cells first observed in each species-dosage combination, and this part of the material could be utilized in a χ^2-analysis. For *T. monococcum* the numbers obtained were at 5000 r: 136, at 10000 r: 390, at 15000 r: 832, the total being 1358. The hypothesis to be tested was that the rate of disturbances (in this case measured by their absolute number) was directly proportional to the X-ray dosage, i. e. the numbers of disturbed divisions should increase in the proportion 1 : 2 : 3. By this hypothesis the 1358 disturbed cells should occur in the following series: at 5000 r: $^1/_6 \times 1358 = 226$, at 10000 r: $^2/_6 \times 1358 = 453$, at 15000 r: $^3/_6 \times 1358 = 679$. As usual in a χ^2-analysis the differences ($-90, -63, +153$), are squared, the squares individually divided by the corresponding values expected, and the quotients ($35{,}84 + 8{,}76 + 34{,}48$) are added to give $\chi^2 = 79{,}08$. The common tables of χ^2 (FISHER, 1936; FISHER and YATES, 1938; BONNIER and TEDIN, 1940) show that with 2 degrees of freedom (number of dosages less one) χ^2 will obtain the value of 13,82 or higher only in one case out of 1000, if the hypothesis tested is true. The value obtained may therefore be said to indicate, with a very high degree of significance, that in *T. monococcum* the rate of disturbed cell divisions does not increase in direct proportion to the dosage of X-rays.

In Table 2 the following data are given: the number of disturbed cells observed and numbers to be expected for each species and dosage,

TABLE 2. *Number of disturbed cells observed and numbers expected in different groups of the material upon the hypothesis of direct proportionality to dosage; χ^2-values for different groups of material.*

Material	Number of mitotic disturbances obtained			Number of mitotic disturbances expected			Σ	χ^2
	5000 r	10000 r	15000 r	5000 r	10000 r	15000 r		
Triticum monococcum	136	390	832	226	453	679	1358	79,08
Triticum durum	245	703	996	324	648	972	1944	24,52
Triticum dicoccum ...	291	860	1041	365	731	1096	2192	40,52
Triticum vulgare	346	737	1004	348	696	1043	2087	3,89
Avena sativa	237	502	828	261	522	784	1567	5,45
Total 4x.....	536	1563	2037	689	1379	2068	4136	58,99
Total 6x.....	583	1239	1832	609	1218	1827	3654	1,48
Total 2x + 4x	672	1953	2869	916	1831	2747	5494	78,55
Total 2x + 6x	719	1629	2664	835	1671	2506	5012	27,13
Total 4x + 6x	1119	2802	3869	1298	2597	3895	7790	41,03
Sum total	1255	3192	4701	1525	3049	4574	9148	58,04

these values being calculated in the manner described in detail above; the value of χ^2 for each single species; the numbers expected and the χ^2 obtained for different combination groups of the material. In the latter cases χ^2 is always calculated from the values for the combination, i. e. the values in the same row of the table. It should be observed that in each case, the value of χ^2 being calculated from 3 groups, the number of the degrees of freedom is 2.

Using the tables of χ^2, we find that neither the diploid nor the tetraploid species agree with the hypothesis of direct proportionality between dosage and rate of disturbances, the significance in each case being very high, whereas there is nothing to indicate that this hypothesis should not be true in the case of the two hexaploid species.

Further, the data may be used to demonstrate the fact that all three degrees of polyploidy differ from one another in regard to the relation between dosage and rate of disturbed cells.

If we first add the 5 χ^2-values from the 5 species, the result is 153,46, corresponding to $5 \times 2 = 10$ degrees of freedom. For the sum total χ^2 is $= 58{,}04$ with 2 degrees of freedom (DF). The difference is 95,42 with 8 DF. This value, being highly significant, tells us that there is heterogeneity in the material, i. e. the different species have different relations to the hypothesis tested. The analysis may be carried further.

If we take the three χ^2-values, corresponding each to a certain degree of polyploidy, their sum is 139,55 with 6 DF, and the difference from the value for the sum total is now 81,51 with 4 DF, again highly significant. In this latter measure of heterogeneity the value of χ^2 was 13,91 less and the DF 4 less than in the one just given. This residue pertains to the differences in proportion, eventually to be found between the two tetraploid species and between the two hexaploids.

The sum of χ^2:s for the two tetraploids, less the χ^2 for the sum of tetraploids, is 6,05, with $4 - 2 = 2$ DF. The value, corresponding closely to the figure for 0,05 in the χ^2-table, is barely significant, and indicates, but does not prove, that the relation of disturbances to dosage is different in *T. durum* and *dicoccum*. For the two hexaploids the corresponding values are 7,86 and 2 DF. This time the value of χ^2 corresponds to $P = 0{,}02$, and it must be considered as strongly indicated that the two species are different in the relation under discussion. None of them could be proved to differ from the straight-line hypothesis, but this must not be taken as a proof that they agree completely with this hypothesis. The fairly high probability that they are different indicates that both of them do not confirm with the hypothesis.

Having thus demonstrated that species of different degrees of polyploidy have different relations between dosage and rate of disturbed cells, we may still ask the question whether all three groups are different, or if two of them show the same relationship. The method, used below, to find an answer to this question, must be used discriminately. When, as in this case, the general measure of heterogeneity has given a very high degree of significance, the method may be safely used.

The sum of χ^2:s for diploid and tetraploids is 138,07, the χ^2 for the sum of these groups is 78,55. The difference, 59,52 with 2 DF is highly significant, proving that there is a difference between those two groups in regard to the relation under discussion. For the difference between diploid and hexaploids the corresponding values are 53,43 and 2 DF, and for the difference 4x versus 6x it is 19,44 and 2 DF. The latter of these values is still highly significant, although of a somewhat lower order of significance than most values hitherto discussed: It is safe to conclude that all three polyploidy-groups differ from one another and that the tetraploids occupy a position somewhere between the diploid and the hexaploids.

In this paper it has thus been conclusively proved that a quantitative change of the chromosome number directly influences the shape of the damage curves appearing after irradiation. The problem

will then arise: Are the two types of curves, the sigmoid and the linear, fundamentally different from one another, or does the last-mentioned type of proportionality only indicate the occurrence of a flattened S-curve? On the other hand, if eventually the latter deduction is true, what significance can be attributed to other instances of a presumed straight-line increase? For a definite proof of the one-hit hypothesis in its various applications, X-chromosome lethals, one-break chromosome aberrations, position effects of *cubitus interruptus* and other genes and, finally, individual gene-mutations must be more thoroughly examined in regard to their dosage relationships at very low as well as middle-high and high dosages than has been the case so far.

SUMMARY.

1. The data published in a previous paper (FRÖIER, GELIN and GUSTAFSSON, 1941) and shown in Table 1 have been submitted to a statistical analysis. The methods used seem to be of value for research work in the field covered and are therefore presented in some detail.

2. The statistical analysis has shown that diploid, tetraploid and hexaploid species respond to increasing X-ray dosages by damage curves, different for each of the three groups of polyploidy. The curves of the single diploid examined and of the two tetraploids deviate significantly from a straight-line proportionality, whereas no deviation from the straight-line can be proved in the case of the hexaploids. Whether the sigmoid and the straight-line curves are fundamentally different cannot be ascertained, but most probably the damage curves of the hexaploids are nothing but flattened sigmoid curves. Instances of straight-line proportionality, inferred from genetical and cytological experiments, must be reinvestigated, special attention being paid to the state of things in low dosage regions (in respect to X-chromosome lethals, for instance, to dosages between 50 and 800 r).

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DIE FAKTORENVERTEILUNG DER LUTESCENS-SIPPE VON AVENA SATIVA AUF DIE ELTERNSORTEN

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(With a Summary in English)

VOR mehreren Jahren fand ÅKERMAN in Nachkommenschaften von Kreuzungen zwischen Abeds Novahafer und Schwarzhafersorten des sogenannten mittelschwedischen Typus eine chlorophylldefekte Hafersippe, f. *lutescens*. In diesen Kreuzungen wurde das Vorhandensein von drei homologen, polymeren Faktoren für den *lutescens*-Typus festgestellt. In der Abhandlung vom Jahre 1922, wo ÅKERMAN über seine Untersuchungen dieser Sippe berichtete, war die Analyse der verschiedenen Faktoren noch nicht so weit geführt, dass man feststellen konnte, wie sich die Faktoren auf die Elternsorten verteilen. Infolge der heller grünen Farbe des Novahafers wurde von ÅKERMAN (1922) die Vermutung ausgesprochen, dass dieser möglicherweise einfaktorig sei (S. 163 und 176). Dies hat sich später allerdings in den weitergeführten Kreuzungsuntersuchungen als nicht richtig gezeigt; der Novahafer besitzt zwei Faktoren für die *lutescens*-Eigenschaft, die mittelschwedischen Schwarzhafersorten dagegen nur einen. Preliminär wurde diese Feststellung schon früher erwähnt (ÅKERMAN, 1929, S. 598) sowie die durch diese Feststellung eingeführten Faktorenbezeichnungen L_n, L_p und L_s (ÅKERMAN und FRÖIER, 1941, S. 398).

Bei der in Rede stehenden Analyse wurden Rückkreuzungen mit einfaktorigen *lutescens*-Heterozygoten ausgeführt. Diese können nämlich auf einem gewissen Entwicklungsstadium von den homozygot normalgrünen Pflanzen mehr oder weniger deutlich unterschieden werden. Wenn sie in ihrer Entwicklung so weit gekommen sind, dass sie 3—4 Blätter haben, treten in den Blättern gelbliche Flecken auf, die bei den Homozygoten dagegen nicht oder nur sehr undeutlich vorkommen (ÅKERMAN, 1922, S. 159). Wenn man auf diesem Stadium aufmerksam das Geflecktwerden der Heterozygoten beobachtet — die Fleckung ist nur während 8—14 Tage ersichtlich, wonach die Blätter wieder vollkommen normal grün werden — kann man in den aller-

meisten Fällen mit Sicherheit die Heterozygoten markieren, um dieselben zu Kreuzungen heranzuziehen. Die Mutterpflanzen werden indessen immer auf ihren Heterozygotencharakter geprüft (Tabelle 1).

Wird eine einfaktorige Sippe mit einer solchen einfaktorigen Heterozygote gekreuzt, hat man ja in F_1 teils normale, grüne Pflanzen und teils gelbgefleckte Heterozygoten in derselben Anzahl zu erwarten, während eine zweifaktorige Sippe bei derselben Kreuzung nur grüne Pflanzen in F_1 ergibt. In der Kreuzung mit einer einfaktorigen Sippe hat man zwei Alternative zu beachten: entweder besitzt die fragliche Sippe denselben Faktor wie die einfaktorige Heterozygote oder nicht. Die zwei Möglichkeiten sind also (mit Faktorsbezeichnungen geschrieben):

A) Eltern: $L_sL_s\,l_pl_p\,l_nl_n \times L_sl_s\,l_pl_p\,l_nl_n$

F_1: 50 % $L_sL_s\,l_pl_p\,l_nl_n$ + 50 % $L_sl_s\,l_pl_p\,l_nl_n$

	Normalgrün, nicht weiter spaltend.	Gefleckt.
F_2:	Normalgrün.	25 % grün, konstant. 50 % gefleckt, weiter spaltend in 3 : 1. 25 % vergilbende *lutescens*-Homozygoten.

B) Eltern: z. B. $l_sl_s\,L_pL_p\,l_nl_n \times L_sl_s\,l_pl_p\,l_nl_n$

F_1: 50 % $L_sl_s\,L_pl_p\,l_nl_n$ + 50 % $l_sl_s\,L_pl_p\,l_nl_n$

	Normalgrün.	Gefleckt.
F_2:	Spaltet in vergilbende, gefleckte und grüne Pfl. nach dem Schema 1 : 4 : 11.	Spaltet in vergilbende, gefleckte und grüne Pfl. nach 1 : 2 : 1.

Bei den zweifaktorigen Heterozygoten (in jeder möglichen Kombination) treten gelbliche Flecken nicht auf. Daher sind auch 50 % von der F_1 (in B) normalgrün.

Die Resultate von Kreuzungen zwischen einfaktorigen Heterozygoten und Glockenhafer II sind in Tabelle 1 zusammengestellt. Wie aus dieser hervorgeht, wurde in F_1 eine Spaltung in grüne und gelbgefleckte in genau dem erwarteten Zahlenverhältnis gefunden.

In der Tabelle 1 fehlen Nachkommenschaften der Mutterpflanzen von den Kreuzungsnummern 208, 209 und 220. Weil aber diese Kreuzungsnummern in F_1 gelbgefleckte Pflanzen gegeben haben, müssen sie unzweideutig aus einfaktorigen Heterozygoten-Mutterpflanzen stammen. Falls die Mutterpflanzen hier grüne Homozygoten gewesen wären,

TABELLE 1. *Kreuzungen einfaktoriger, gelbgefleckter Heterozygoten mit Glockenhafer II.*

Feld-nummer 1923	Kreuzungs-nummer 1922	Spaltung der Nachkommenschaften der Mutterpflanzen			Spaltung in F_1		Bemerkungen
		Grün	Gelbgefleckt	*lutescens*	Grün	Gelbgefleckt	
907_1	165 ♀	10	21	10	—	—	Die gelbgefleckten Mutterpflanzen in der Reihe 1923—907 stammten aus der Kreuzung Novahafer × Glocken III, diejenigen in der Reihe 1923—908 aus der Kreuzung Novahafer × Glocken II.
2	»	—	—	—	—	1	
3	172 ♀	32	85	41	—	—	
4	»	—	—	—	1	1	
9	176 ♀	7	38	18	—	—	
10	»	—	—	—	—	—	
11	177 ♀	3	25	12	—	—	
12	»	—	—	—	1	—	
17	182 ♀	10	26	20	—	—	
18	»	—	—	—	—	1	
Summe		62	195	101	2	3	
Erwartet.....		89,5	179,0	89,5	2,5	2,5	
908_1	190 ♀	5	20	14	—	—	
2	»	—	—	—	—	1	
6	199 ♀	8	10	5	—	—	
7	»	—	—	—	1	1	
15	208	—	—	—	—	1	
16	209	—	—	—	—	1	
17	210 ♀	4	4	1	—	—	
18	»	—	—	—	5	—	
19	211 ♀	5	5	2	—	—	
20	»	—	—	—	1	2	
26	220	—	—	—	1	1	
Summe		22	39	22	8	7	
Erwartet.....		20,8	41,4	20,8	7,5	7,5	
Summe sämtlicher Nummer:		84	234	123	10	10	
Erwartet:		110,3	220,4	110,3	10,0	10,0	

fälschlich als gefleckte Heterozygoten angesehen und markiert worden, hätten ja in F_1 keine gefleckten Pflanzen auftreten können.

Aus der Tabelle 1 kann man also den Schluss ziehen, dass der Glockenhafer II in bezug auf die *lutescens*-Eigenschaft einfaktorig sein muss. In der Tabelle sind ebenfalls die Spaltungszahlen, welche in den Nachkommenschaften der Mutterpflanzen erhalten wurden, aufgeführt. Wie daraus ersichtlich, ist die Anzahl der grünen etwas zu klein, was ohne Zweifel damit zusammenhängt, dass einige von diesen als gelbgefleckte Heterozygoten klassifiziert worden sind. ÅKERMAN hatte selbst keine Möglichkeit, diese Klassifizierung zu kontrollieren, dagegen hat

TABELLE 2. *Kreuzungen einfaktoriger, gelbgefleckter Heterozygoten mit Abeds Novahafer.*

Feld-nummer 1923	Kreuzungs-nummer 1922	Spaltung der Nachkommenschaften der Mutterpflanzen			Spaltung in F_1		Bemerkungen
		Grün	Gelbgefleckt	*lutescens*	Grün	Gelbgefleckt	
907_5	173 ♀	3	7	3	—	—	Vergleiche Tabelle 1 bezüglich Abstammung der gelbgefleckten Mutterpflanzen!
6	»	—	—	—	3	—	
7	174 ♀	3	25	10	—	—	
8	»	—	—	—	1	—	
13	179 ♀	13	25	12	—	—	
14	»	—	—	—	2	—	
15	181 ♀	5	13	2	—	—	
16	»	—	—	—	5	—	
Summe		24	70	27	11	—	
Erwartet.....		30,3	60,4	30,3	11,0	—	
908_3	193	—	—	—	3	—	[1] Die Mutterpflanze unrichtig klassifiziert. In den Durchschnittszahlen nicht mit einberechnet.
8	201	—	—	—	1	—	
9	202	—	—	—	1	—	
10	203	—	—	—	1	—	
11	205 ♀	(50)[1]	—	—	—	—	
12	»	—	—	—	(2)[1]	—	
13	206 ♀	6	17	9	—	—	
14	»	—	—	—	3	—	
15	213	—	—	—	1	—	
Summe		6	17	9	10	—	
Erwartet.....		8,0	16,0	8,0	10,0	—	
Summe sämtlicher Nummer:		30	87	36	21	—	
Erwartet:		38,3	76,4	38,3	21,0	—	

er die von den F_1-Pflanzen, die sehr genau mehrmals während der »kritischen» Zeit von ihm beobachtet wurden, kontrolliert. Die Zahl der *lutescens*-Pflanzen ist etwas grösser als die erwartete; in einer χ^2-Analyse, wo grüne und gelbgefleckte Pflanzen zusammengenommen werden, erhält man $\chi^2 = 1{,}950$ und $0{,}20 > P > 0{,}10$. In 10 bis 20 % aller Fälle von einer ähnlichen Spaltung könnte also eine Verteilung wie die erhaltene auftreten.

Die Resultate von Kreuzungen zwischen einfaktorigen Heterozygoten und Abeds Novahafer sind in Tabelle 2 zusammengeführt worden. Wie aus dieser hervorgeht, wurden in F_1 nur grüne Pflanzen erhalten, was also die Befunde bestätigt, dass der Novahafer unter den beiden fraglichen Elternsorten die zweifaktorige ist.

Die Mutterpflanzen der Kreuzungsnummer 193, 201, 202, 203 und 213 waren so unzweideutig gefleckte Heterozygoten, dass eine Kontrollprüfung hier nicht notwendig war. Die Mutterpflanze 908_{11} (= 205 ♀) war schon 1922 als »grün oder gelbgefleckt?» bezeichnet, was ja auch im Einklang mit dem Befunde ihrer nichtspaltenden, homozygoten Natur im Jahre 1923 steht.

In ÅKERMANS Arbeit vom Jahre 1922 wurde nur über Kreuzungen zwischen Novahafer und den drei Schwarzhafersorten Glockenhafer II, III und Grossmogulhafer berichtet. Da in sämtlichen diesen Kreuzungen eine Spaltung von grünen : *lutescens*-Pflanzen in dem Zahlenverhältnis 63 : 1 gefunden wurde, müssen alle diese Sorten einfaktorig sein. Dieser Faktor, den genannten mittelschwedischen Hafersorten eigen, bezeichnen wir als L_s. Einfaktorige Heterozygoten von der Formel $L_s l_s\ l_p l_p\ l_n l_n$ sind später von FRÖIER (unveröffentlicht) durch Kreuzungen von gelbgefleckt × Glocken II isoliert worden; hierbei wurde in F_1 eine Spaltung in grün: gefleckt nach 1 : 1 erhalten, und die gefleckten spalteten genau nach 3 : 1, so wie das Schema A, Seite 172, fordert; die grünen bleiben konstant. Mit dieser Heterozygote werden z. Zt. eine Anzahl der phylogenetisch interessantesten Hafertypen von FRÖIER gekreuzt. Unter anderem werden alte skandinavische Landsorten sowie Probsteiersorten verschiedener Herkunft analysiert, um eventuelle verschiedene Faktorenverteilungen zu entschleiern, woraus Schlüsse über unsichtbar verlaufene Mutationsschritte gezogen werden können. Die Faktorenverteilung des Novahafers ist $l_s l_s\ L_p L_p\ L_n L_n$, wo L_p den dem Novahafer und den bisher untersuchten Probsteierstämmen gemeinsamen Faktor darstellt. In der 1929-er Arbeit von ÅKERMAN sind eine Reihe Probsteiersorten aufgeführt worden, die nach Kreuzung mit Abeds Novahafer keine *lutescens*-Spaltungen ergeben. Als L_n endlich wird der dritte Faktor bezeichnet, der sich in Abeds Nova befinden muss — ob auch in übrigen untersuchten Probsteiersorten, die dann dreifaktorig sein müssen ($L_s L_s\ L_p L_p\ L_n L_n$), liegt im Untersuchungsrahmen der von FRÖIER fortgesetzten Kreuzungsstudien.

SUMMARY.

In his study of the genetics of the remarkable f. *lutescens* of cultivated oats, the senior author showed that the segregation of *lutescens* homozygotes was brought about by three homologous, polymeric factors, showing free combination and producing green colour in the heterozygous state (ÅKERMAN, 1922). The distribution of the different

factors in the parental types, Abeds Nova oats (Danish Probsteier strain) and middle-Swedish black-hulled oats was not then determined. The suggestion was made (English Summary, p. 176, l. c.) that the light green colour of the Nova oats might be connected with the presence of only one chlorophyll factor in this type. The prediction, however, failed. In crossing monofactorial, »yellow-stained» heterozygotes with Klock II (Bell II), Table 1 shows, on the contrary, the agreement with the hypothesis of the black-hulled oat parent possessing only one factor, F_1 giving 50 per cent normal green plants and 50 per cent »yellow-stained» (bifactorial heterozygotes are normal green without any yellow dots on the leaves which are characteristic of the three possible monofactorial heterozygotes). Similarly, Table 2 shows the results from crosses with monofactorial heterozygotes $\times$ Abeds Nova oats, proving the bifactorial constitution of the latter parental strain: no »yellow-stained», monofactorial heterozygotes appeared in F_1 which should have happened if Abeds Nova possessed only one *lutescens* factor.

The distribution of the three factors, L_s, L_p and L_n, is discussed. Further crosses are being made by the junior author (FRÖIER; unpublished) in order to reveal the factorial distribution in old Scandinavian land oat strains, and also in Probsteier oats of different origin.

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2. — 1929. Letalfaktorer hos havre och vete. — Beretning om Nordiske Jordbrugsforskeres Forenings IV. Kongres, Helsingfors. S. 594—604.
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STUDIES ON THE MEIOTIC MECHANISM OF HAPLOID RYE

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MEIOSIS is adapted to diploid organisms and acts within them with precision. In polyploid or polysomatic forms deviations from the normal meiotic course occur and result in more or less unbalanced conditions in the gametes. The study of these meiotic variations has increased our knowledge of the factors underlying the normal meiotic process. Thus, the main difference between mitosis and meiosis can be ascribed to the association in pairs of homologous chromosomes during early meiotic prophase. Even if more than two homologous chromosomes of each kind are present only pairs of chromosomes attract each other. This tendency of the smallest parts of the chromosomes to exist in pairs during meiotic prophase is so strong that if no partner for a certain chromosome exists, it may pair with another non-homologous chromosome, or with another part of itself. Although indications of such non-homologous pairing are not infrequently met with, it has been analysed in detail only in maize (McCLINTOCK, 1933). It is still unknown whether non-homologous pairing is essentially different from ordinary pairing. It is sometimes described as »ineffective pairing», indicating that it is probably less intimate than normal pairing and above all that it should not be able to cause chiasma formation.

Chromosomes without homologous partners occur in all triploids, trisomics, etc. But their maximal occurrence is realized in the monohaploid organism, where all the chromosomes lack partners. For this reason it is of the outmost importance to study meiosis in haploids. Several such investigations have been made. Owing to the scanty occurrence of the haploids, however, it has not been possible to choose the material from among cytologically favourable plants. And of the available haploids only few can be kept alive long enough to permit a study of their meiosis. Further, even if meiosis can be fixed, the inferior viability of the plants very often makes the fixations less suitable. As a result most haploids in which meiosis has been studied have not been suited for a critical analysis of the different meiotic stages.

Therefore, the greater part of the observations of haploid meiosis has often been confined to the mere establishment of numerous disturbances during the first and second division, causing an almost absolute gamete sterility. Up to the present the prostages, for instance, which from a meiotic-mechanical point of view are of critical significance, have in no case been followed completely from zygotene to the disappearance of the nuclear membrane at metakinesis.

I was fortunate in obtaining one haploid twin plant of rye in the winter of 1940—41. Having previously noted that rye is in many respects an ideal cytological material, I decided to try to analyse the course of meiosis as closely as possible in this haploid. Already a preliminary study showed that the prostages in particular ran a rather remarkable course. I found a typical pachytene, in which the chromosome length present exhibited up to 100 per cent pairing. At diplotene and diakinesis maximal pairing was sometimes found, all the seven chromosomes being joined into two bivalents and one trivalent. Even at late diakinesis this extreme pairing could be traced, while numerous chiasmata kept some of the paired chromosomes together at metaphase I.

Through the courtesy of Professor MÜNTZING three other haploid rye plants, spontaneously originating in his population material, were handed over to me. Meiosis could be studied in two of these plants, while the third plant only yielded fixations of somatic chromosomes. The two first-mentioned plants, however, showed the same characteristic peculiarities in their meiotic prophase as those met with in the first plant, although the number of persisting chiasmata was considerably less in one of them. Thus, the following analysis of meiosis of haploid rye is based upon three individuals:

443 b. Twin plant from population rye. The other twin was a normal diploid. This haploid, although of course considerably weaker than normal diploids, was relatively strong. It had many culms about 60 cm in height shooting at intervals. In all, 7 ears were formed. The leaf breadth was 7 mm. Chiasma frequency 0,65 per cell.

317—26. Spontaneous haploid population plant. This plant was also very vigorous, even somewhat more robust than 443 b. It formed 7 ears, the highest culm being 55 cm, and the leaf breadth 7 mm. Chiasma frequency 0,09 per cell.

343—10. Also a spontaneous population haploid, decidedly weaker than the two preceding plants. It formed one vigorous culm, 53 cm

in height, and one weak culm. Leaf breadth 5 mm, chiasma frequency 0,72 per cell.

In all, 9 fixations were made, from which some 30 usable slides resulted. Although several of the fixations turned out inferior, as is usually the case in haploids, I succeeded in getting some first-rate fixations of all stages. NAVASHIN was used as fixative, LEWITSKY also being tried for the root chromosomes. The staining in gentian violet was supplemented with FEULGEN stainings, and nucleolus staining in light-green according to BHADURI.

I am very much indebted to Professor A. MÜNTZING, Lund, for supplying material, to Dr. O. TEDIN, Svalöf, for the statistical analysis of chiasma frequencies, and to Miss MAGNA PALM, Svalöf, for making the slides for this investigation.

I. THE SOMATIC CHROMOSOMES.

Root chromosomes were studied in all the four haploids available and also in two haploid rye plants previously detected at this laboratory (MÜNTZING, 1937; NORDENSKIÖLD, 1939). They all showed the normal type of rye idiogram; aberrant chromosomes, reported on several earlier occasions in *Secale*, were not met with in these haploids.

Fig. 1 *a* shows a root mitosis of haploid rye. The centromeres are more or less medially located. Two of the chromosomes, however, have rather asymmetric arms. One of them, in the continuation called s_1, is provided with a large satellite on its shorter arm, and the nucleolus is organized by the achromatic portion that separates the satellite from the chromosome (Fig. 1 *e—g*). This chromosome is the only satellite carrier which may be easily recognized in untreated diploid rye. It has been described on numerous occasions both in root chromosomes (for instance, LEWITSKY, 1931; MÜNTZING, 1937; PATHAK, 1940) and in the pollen mitosis (HASEGAWA, 1934). It is clearly seen also during the second meiotic division (Fig. 1 *b*). I found its proportions to agree with earlier observations. The mean of five measurements in root mitoses gave the following values: the satellite, 1,2 μ, the short arm 1,9 μ, the long arm 4,0 μ, altogether 7,1 μ.

The other asymmetric chromosome (s_2) is also furnished with a satellite, but this is much more difficult to reveal. In one of the haploids 317—26, however, it was regularly visible (Fig. 1 *a*, *d*, *h—j*). The satellite of s_2 is also attached to the shorter arm of the chromosome. It differs in appearance from the s_1 satellite. While the s_1

satellite is rod-shaped and has the same, or slightly smaller, diameter than the chromosome, the s_2 satellite is ovoid and of a considerably smaller diameter. The dimensions of the s_2 chromosome are the following (means of 5 measurements): the satellite, 0,4 μ, the short arm 1,7 μ, the long arm 4,4 μ, total 6,5 μ.

In 317—26, the same plant as that in which s_2 was detected, there often, although more irregularly, occurred another satellited chromo-

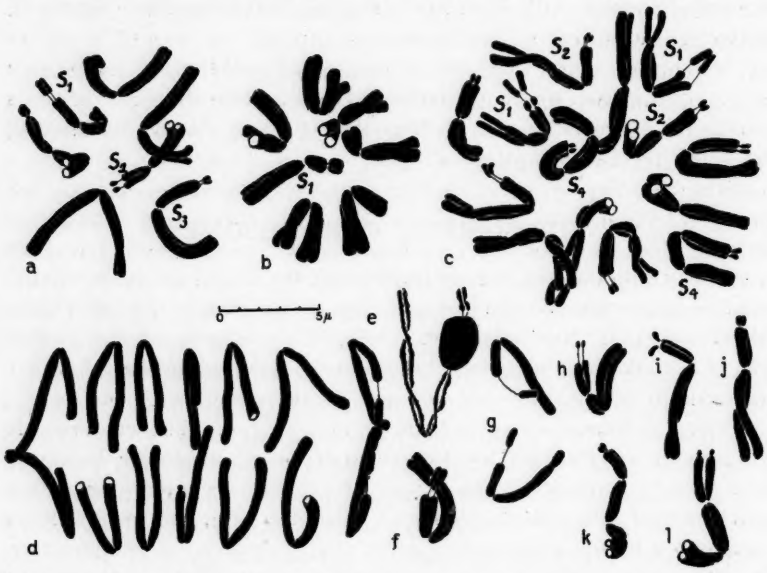

Fig. 1. *a*: haploid root metaphase, *b*: second meiotic metaphase in the haploid, *c*: diploid root metaphase, chromosomes treated with cold during 3 days, *d*: haploid somatic anaphase, *e—g*: instances of s_1, *h—j*: s_2, *k—l*: s_4. — × 3000.

some, called s_3 (Fig. 1 *a*). This is evidently one of the shorter submedially inserted chromosomes. In the same plant still more satellites could be seen now and then. These observations, and the fact that also in diploid population plants other satellites than s_1 were occasionally encountered, encouraged me to arrange a few experiments with cold treatments in order to make these differentiations more evident. Although these attempts were made on a very small scale, positive results were obtained. Owing to the preliminary character of the experiments the results should not, however, be generalised.

25 kernels of the commercial variety »Steel rye» were put to

germinate on filter paper at room temperature. After 2 days, when root tips began to appear, they were placed in a thermostat at 0° C. One third of the kernels was fixed after a cold treatment of 1, 3 and 5 days respectively. It was found that in most of the chromosomes the shorter arm had differentiated into a longer interior portion of normal diameter and a shorter terminal portion decidedly thinner and less stained. Already one day's cold treatment brought about a regular appearance of the s_2 chromosome with a fully developed satellite, while 3 days in cold resulted in such pictures as Fig. 1 *c*, in which 10 of the 14 chromosomes were clearly differentiated. After a treatment of 5 days the spindle was often put out of function and exactly the same effect was obtained as in colchicine treatment. Thus, at metaphase typical c-pairs were found irregularly scattered over the cell. This is hardly surprising, as cold treatment for a long time has been known to induce chromosome doublings. The chromosome plate in Fig. 1 *c* was fixed in NAVASHIN and stained in FEULGEN. A similar result is obtained by fixing in LEWITSKY and staining in gentian violet. The slides showed that, in addition to s_1 and s_2, evidently 3 pairs of chromosomes exhibited satellite-like differentiations. s_3 is probably one of these. Another pair, s_4 (Fig. 1 *k*—*l*), had a clear achromatic portion in the middle of the long arm and seems to correspond to the B-chromosome of KATTERMANN (1937, 1938) and to the chromosomes participating in an amphibivalent, which MÜNTZING and PRAKKEN (1941, Figs. 80—111, and unpublished) found to have a similar constriction. Four of the longest chromosomes are still undifferentiated after a treatment of 5 days.

The works of DARLINGTON and LACOUR (1940, 1941) have shown that these differential reactions of the chromosomes, which are obtained by cold treatment, are really due to the different distribution of euchromatin and heterochromatin along the chromosomes. The heterochromatic regions are deprived of their nucleic acid during the treatment and appear after that as narrower and lighter sections in the chromosomes. Cold treatment brings about a kind of »nucleic acid starvation». It may consequently be assumed that the rye chromosomes have heterochromatic end-segments in their shorter arms. This is in agreement with a condition found at pachytene, viz. terminal knobs occurring in several of the chromosome pairs.

A similar marked uniformity in the position of the heterochromatic portions has been demonstrated also in the other plants, where cold treatment has been successful in revealing the heterochromatin. In

Fritillaria (DARLINGTON and LACOUR, 1941) the heterochromatin is proximally localised in several bands close to the centromeres, in *Paris* and *Trillium* (DARLINGTON and LACOUR, 1938, 1940) and in *Adoxa* (GEITLER, 1940 a) the localisation is distal, as in *Secale*. KOSTOFF, using another technique, has found heterochromatic regions distally located in the chromosomes of *Crepis capillaris* and *Triticum monococcum* (1938 a, c). LONGLEY (1939, 1940), working with *Zea* and *Euchlaena*, points out the regularity in position of the pachytene knobs: in *Zea* their position is almost throughout interstitial, and it has been shown that a certain distance from the centromere gives the optimal conditions for their development; in *Euchlaena*, on the other hand, all the knobs are terminal.

It is of interest to note that some of these differentiations, which in diploid rye are found only after cold treatment, may nevertheless occur normally in one of the haploid plants. The lower viability of the haploid evidently acts in the same manner as cold treatment, in the haploid a certain degree of nucleic acid starvation seems to be the normal condition.

II. THE MEIOTIC PROPHASE.

In very young pollen mother cells, at preleptotene and leptotene, there is present one large nucleolus, stained faintly by gentian violet but remaining unstained by FEULGEN. There also appears very regularly another nucleolus-like body, which is stained deeply both in gentian violet and in FEULGEN, and consequently must consist of nucleic acid (Fig. 2 *a*). Sometimes two, or even three, such bodies may be seen, but they are then smaller in size. These bodies are probably the »fused heterochromatic bodies» which DARLINGTON and LACOUR (1941, Fig. 3) picture in pachytene of the embryo sac mother cell of *Fritillaria pudica*. As the pollen mother cells gradually increase in size, the chromosomes begin to be stainable more clearly. They are then seen to come together in pairs. The pairing seems to start at the terminal knobs, present in several of the chromosomes. These are evidently heterochromatic and may have fused already during the resting stage. The pairing proceeds along the chromosomes and it can be clearly seen that the chromomeres pair. This condition constitutes a striking resemblance to the homologous pairing. Also in the haploid, where most of the pairing must be non-homologous, there is evidently an attraction between pairs of chromomeres. The lack in homology

within the chromomere pairs is often visible as a morphological difference in size or shape, but usually no such difference is detectable.

The pairing continues for a long time and in certain instances it becomes almost complete. A stage is reached which closely resembles ordinary diploid pachytene. A close study, however, always reveals numerous irregularities. I am not sure that this stage is always attained, as in many cases the pairing seems to be interrupted before being

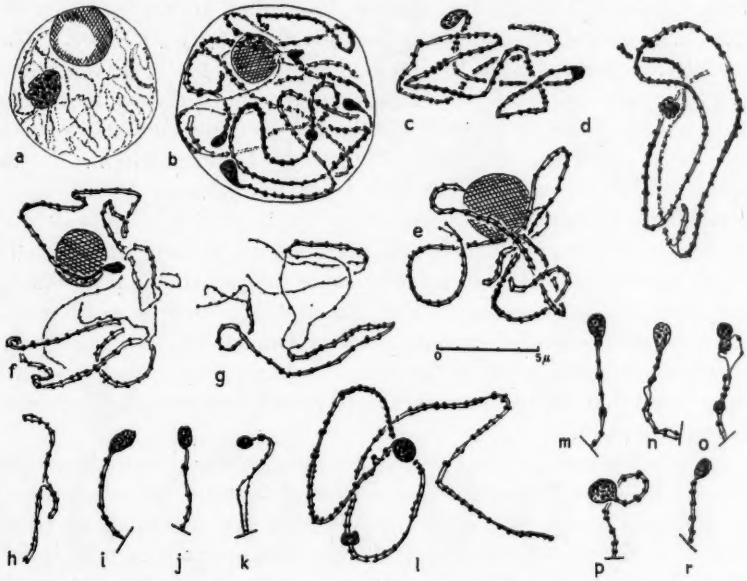

Fig. 2. *a*: haploid leptotene, *b*: general appearance of haploid pachytene, *c*—*h*: instances of haploid pachytene configurations, *i*—*k*: terminal segments of haploid pachytene bivalents, *l*: diploid pachytene bivalent, *m*—*r*: terminal segments of diploid pachytene bivalents. — × 3100.

completed. This is the case especially in those cells which lie in the middle of the pollen sac without contact with the tapetum. These cells are always greatly retarded in their development. Only in one fixation were all chromosomes nearly completely paired, otherwise the pairing could generally be estimated at 50—80 % of the chromosome length.

Nor can I decide for certain whether the two plants with a high chiasma frequency had a more complete pachytene pairing than the third plant, which at metaphase had only a few chiasmata. Most slides did not show any correlation in this respect. This is perhaps

not astonishing, since the asynaptic *Allium amplectens* (LEVAN, 1940) had just as intimate a pachytene pairing as other *Allium* forms, although no chiasmata at all were formed. It is impossible to decide from the appearance of the pachytene pairing if chiasmata will be formed.

Pachytene nuclei of the haploids are undoubtedly more difficult to analyse completely than the corresponding stage in diploid rye. This is the case also in *Zea* (MCCLINTOCK, 1933) and *Antirrhinum* (ERNST, 1939), where the diploid pachytene has been fully analysed. This condition is due partly to the smaller volume of the haploid nucleus, which in spite of the lower chromosome number results in a great crowding, and partly to the numerous anomalies of the pairing, thus often rendering the situation difficult of interpretation. It was established, however, with certainty that a surprisingly large number of normally paired regions occurred in the haploid rye. It was often possible to follow entire pachytene bivalents along their whole length. It was repeatedly observed that the ends of the two components of each pair were on the same level. This observation, which at pachytene is necessarily somewhat uncertain, is in agreement with the appearance of the chromosome pairs at later stages. Especially at diakinesis it is often noted that the chromosomes have paired with the same regularity as in the diploid.

The agreement in level within the pairs indicates that the pairing starts at the ends. I therefore devoted some attention to the appearance of the chromosome ends both in the diploid and the haploid. In the diploid 4 or 5 of the 14 ends terminate with an ovoid or pear-shaped knob (Fig. 2 *l*—*r*). These terminal knobs are also present in the haploid (Fig. 2 *b*, *c*, *i*—*k*). They are smaller than in the diploid, but they have a similar appearance and staining qualities. As has already been pointed out, these lumps probably correspond to the heterochromatic end segments of the somatic chromosomes. At pachytene, however, some intercalary knobs were also found.

Fig. 2 *c*—*e* pictures three whole pachytene bivalents. They are almost completely paired and their pairing is strikingly regular. In Fig. 2 *d* a clear foldback is seen at the bottom. In Fig. 2 *f*—*g* several unpaired regions are present, and in *f* an exchange of threads occurs between different pairs, which may result in multivalent formation. Fig. 2 *h* is another example of a foldback.

At the transition to diplotene the chromosomes shorten considerably and the different configurations are now more easily understood.

The bivalents may be seen to develop an equal relational spiral (Fig. 3 *a*). At a somewhat later diplotene whole cells can be readily analysed. It is now evident that the main part of the pairing consists of a spiralisation pairing without chiasmata. As this type of pairing is a gradually disappearing rest of the earlier pairing it may be called a *residue pairing*, in contradistinction to the pairing by chiasmata.

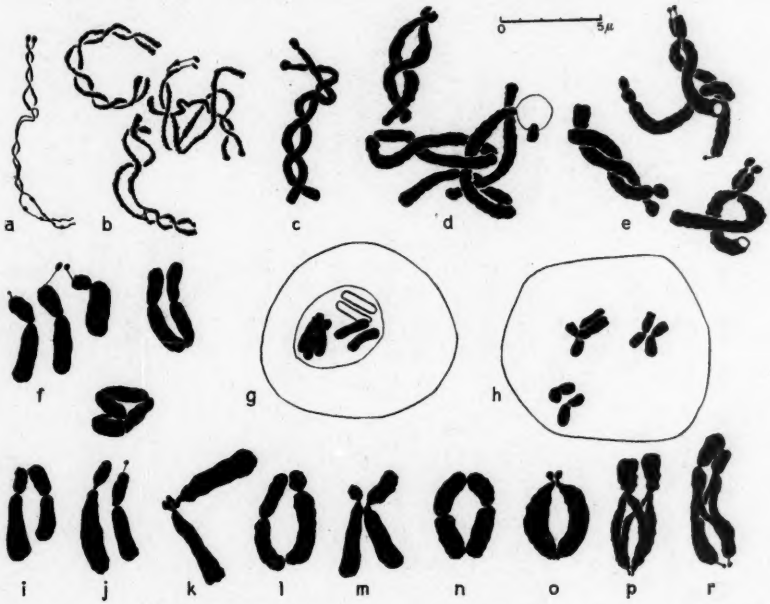

Fig. 3. *a*: a bivalent at early diplotene, *c*: at late diplotene, *b*: the chromosomes of a whole cell at diplotene, *d*—*e*: early diakinesis, *f*: diakinesis, *g*, *h*: the transition to metaphase I, the chromosomes clearly show residue pairing, *i*—*r*: instances of diakinesis configurations. — *a*—*f*, *i*—*r*: × 3000, *g*—*h*: × 1500.

The degree of residue pairing differs widely in different cells. The cell shown in Fig. 3 *b*, with its logical continuation in Fig. 3 *d*, *e*, *f* and Fig. 4 *e*, has a maximal pairing. Its chromosomes form two bivalents and one trivalent. Most of its pairing, however, is residue pairing, only one real chiasma could be detected. It may be pointed out that although no other type of diplotene configurations is pictured, the type shown in Fig. 3 *b*, with complete trivalent and bivalent pairing, is by no means the commonest type. Already at diplotene one or two univalents have usually separated from the paired complexes, and in extreme cases

whole cells may contain exclusively univalents with only a few loops left of the residue pairing. The configurations of the reproduced cell are also more schematic than typically. Frequently all the chromosomes are coiled together in very complex and intricate spirals. The bivalent type of Fig. 3 *b* and *c* is very typical, however, with its ends on an equal level and with 5—8 gyres of coiling. One or two such bivalents are found in most of the cells. During this stage it is often

Fig. 4. *a—f*: metaphase I configurations of whole cells, *g*: instances of ring-shaped univalents, *h*: instances of bivalents and multivalents, *i—j*: anaphase I, *k—m*: chromatid bridges at anaphase I, *n*: telophase I, *o*: interkinesis. — × 2200.

seen that the chromosome ends carry satellites. These seem to be paired and they probably form the remainder of the pachytene knobs.

Diakinesis develops from diplotene by continued contraction. At early diakinesis (Fig. 3 *d—e*) there is still present a pronounced residue pairing into bivalents and trivalents, with the chromosomes held together by several spiral coils. But with the gradually increasing contraction the spirals uncoil and at a more advanced diakinesis stage the residue pairing is reduced to a side by side arrangement of the

chromosomes, whereby often only the ends touch each other, while the rest of the two chromosomes are arched (Fig. 3 *f*—*r*). Several of the satellites may be recognised at diakinesis. The s_1 chromosome and its connexion with the nucleolus in particular are often distinct.

It would have been of great importance to ascertain whether the same chromosomes have a tendency to join in pairs, or if the pairing is entirely at random. Unfortunately, this question could not be definitely answered. Very often, however, it was observed that pairing took place between chromosomes with equal-sized satellites and with the centromeres on the same level. If two asymmetric chromosomes paired, the short arm of one component never paired with the long arm of the other chromosome. This undoubtedly indicates a certain regularity in the pairing, a regularity which could be brought about, for instance, if the pairing started at the heterochromatic ends of the short chromosome arms.

It is evident that some of the terminal associations during diakinesis are real chiasmata. In fact, these ring-shaped configurations are very similar to the normal diploid diakinesis bivalents, which certainly have two chiasmata. It is difficult, however, in the haploid diakinesis to distinguish between residue pairing and real chiasmata. And the development into metaphase I shows that only a small number of the chromosomes paired at diakinesis contain chiasmata. Interstitial chiasmata very rarely occur at this stage, and they are directly visible (Fig. 3 *p*, *r*).

The chromosome pairing in bivalents and trivalents was subject to great variation, also between different pollen sacs in the same plant. Evidently quite special conditions are necessary at zygotene, if the pairing is to reach its maximum. Fig. 3 *g* and *h* are two cells from a pollen sac in which all cells showed a very pronounced residue pairing. In *g* the nuclear membrane still remains and the chromosomes are gathered in the centre of the cell (»third contraction»), in *h* the membrane has disappeared and the chromosomes are scattered in the plasm. Even at this metakinesis stage a very evident residue pairing could be seen in almost every cell. And here there was no doubt that most of the pairing was really residue pairing, as the few chiasmata present could be directly recognised. Thus we met here a similar stability in the arrangement of the chromosomes as that encountered in *Allium amplectens,* where at metaphase I the trivalents could be reconstructed from their residue pairing in spite of the total absence of chiasmata.

III. THE FIRST DIVISION.

After the disappearance of the nuclear membrane the chromosomes spread in the cell. The univalents do not react to the spindle, while the bivalents and multivalents immediately take up a regular arrangement between the poles. It is now easy to analyse whole cells. Such an analysis was made in 6 different fixations of the three haploids. The result is summarised in Table 1.

From this table it will be seen that the three plants behave differently in their chiasma formation. 317—26 behaves in the way characteristic of haploids. Usually only univalents occur, now and then a bivalent is found. The frequency of bivalents is about one in every tenth cell. In a certain pollen sac, however, this frequency increased considerably, one bivalent being found in every third cell. This shows that the chiasma formation is susceptible to modificative influences. This has often been pointed out in haploids (e. g. *Godetia*; HÅKANSSON, 1940). In the only case of haploid rye previously studied, the frequency of bivalents was less than in this plant 317—26. It was estimated at about one chiasma in every 30th cell (NORDENSKIÖLD, 1939).

Both the other haploids, 343—10 and 443 b, had about ten times more chiasmata than 317—26. In these plants not only bivalents are present but also trivalents and even on rare occasions quadrivalents. Not less than 10 different combinations of uni-, bi- and multivalents are recorded, corresponding to a chiasma frequency of up to 4 per cell. In Fig. 4 *a—f* are given a few instances of cells with a varying frequency of different configurations.

The univalents closely resemble contracted somatic chromosomes. Their centromeric constriction is usually concealed. It is possible, however, to discern one or two chromosomes, which have asymmetric arms, and therefore must correspond to s_1 and s_2, but their satellites are embodied in the chromosome bulk. In rare cases the univalents may become ring-shaped, owing to a chiasma being formed between the two arms of the univalent. Such ring-shaped univalents were observed 7 times (Fig. 4 *d, g*). In some cases the chiasma is terminal but usually it is located at some distance from the ends. These chiasmata are the result of foldback pairing, which is not infrequently observed at pachytene.

The bivalents are exclusively rods. Only in one doubtful case was a ring-bivalent observed. The bivalents react immediately to the spindle, and locate themselves in its longitudinal direction. Where

TABLE 1. *Metaphase I configurations of haploid Secale.*

Plant No.	Fixation No.	Number of	Configuration No. 1	2	3	4	5	6	7	8	9	10	Total of cells	Total xata	xata/cell
		I	7	5	3	1	4	2	—	1	3	1			
		II	—	1	2	3	—	1	2	—	—	1			
		III	—	—	—	—	1	1	1	2	—	—			
		IV	—	—	—	—	—	—	—	—	1	1			
		xata	0	1	2	3	2	3	4	4	3	4			
343—10	1	Number	76	89	22	1	7	2	—	1	2	—	200	166	0,83
		%	38,0	44,5	11,0	0,5	3,5	1,0	—	0,5	1,0	—			
	2	Number	235	206	37	3	13	3	1	—	—	2	500	336	0,67
		%	47,0	41,2	7,4	0,6	2,6	0,6	0,2	—	—	0,4			
443 b	1	Number	246	257	35	2	11	1	—	—	—	—	552	358	0,65
		%	44,6	46,6	6,3	0,4	2,0	0,2	—	—	—	—			
317—26	1	Number	729	78	2	—	—	—	—	—	—	—	809	82	0,10
		%	90,1	9,6	0,3	—	—	—	—	—	—	—			
	2	Number	460	32	1	1	1	—	—	—	—	—	495	39	0,08
		%	92,9	6,5	0,2	0,2	0,2	—	—	—	—	—			
	3	Number	50	21	1	—	—	—	—	—	—	—	72	23	0,32
		%	69,4	29,2	1,4	—	—	—	—	—	—	—			
NORDENSKIÖLD, 1939 ...		Number	465	15	—	—	—	—	—	—	—	—	480	15	0,03
		%	96,9	3,1	—	—	—	—	—	—	—	—			
Total		Number	2261	698	98	7	32	6	1	1	2	2	3108	1019	0,33
		%	72,75	22,46	3,15	0,23	1,03	0,19	0,03	0,03	0,06	0,06			

more than one bivalent is present in a cell, the bivalents are arranged parallelly. Unfortunately, the chromosomes cannot be identified at metaphase I, so the important question as to whether certain chromosomes build up the bivalents remains unanswered. The only chromosome which can be recognised with some degree of certainty is s_1. In one pollen sac s_1 was found to occur in 12 among 100 bivalents observed. The general appearance of the bivalents is shown in Fig. 4 *b—f*, *h* 1—7. They resemble the ordinary rod bivalents present in diploid rye. Their chiasma is formed with great regularity and at metaphase it is either terminal or located close to the ends. Asymmetric chiasmata are not often seen. On a thorough examination, however, sometimes evidence of such asymmetry is found. Thus, in one case 11 % asymmetric chiasmata were counted. Instances of asymmetric chiasmata are pictured in Fig. 4 *h*, 5, 6, 7. The terminalisation varies somewhat. In one case 70 % of the chiasmata were completely terminalised, while 30 % showed small cross-arms. The terminalisation evidently proceeds during metaphase, and it is impossible to identify any certain bivalent from its degree of terminalisation.

Trivalents occur both combined with only univalents and together with one or two bivalents. Once 2 trivalents occurred in one cell. The trivalents are usually chains, but in some sure cases trivalents with a triple chiasma were observed (Fig. 4 *e*, *h* 8—13). Quadrivalents were recorded in 4 out of 2628 cells. Three of them were chains, one was a branched quadrivalent with a triple chiasma (Fig. 4 *f*, *h* 14—15).

The first sign of anaphase is the becoming visible of the split between the chromatids of the univalents. At the same time the univalents begin to gather towards the poles (Fig. 4 *i*, *j*). Usually the univalents proceed directly to the poles without first forming any equatorial plate. Solitary univalents, however, may lag on the equator and eventually divide. This has happened to the s_1 univalent of Fig. 4 *j*. Sometimes all the univalents may lag on the equator and finally, undivided, form one monad nucleus. This is the prostage of the occasionally observed unicellular second divisions with somatic chromosome number (the *Allium amplectens* type of meiosis). A monad interkinesis occurred in most cases in haploid *Godetia Whitneyi* (HÅKANSSON, 1940) and its development was often preceded by the formation of an equatorial plate consisting of univalents. In rye, on the other hand, a monad interkinesis belongs to the exceptions. In most cases 2 interkinesis nuclei and two cells are found, even if only a single chromosome is present in one of the cells.

The univalents of the first division often assume a telophase appearance before reaching the poles (Fig. 4 *n*), and in that case the interkinesis stage is very often almost eliminated, the chromosomes immediately forming one or two irregular second metaphases. On the whole, it may be said that up to metaphase I the development takes place with a fairly great uniformity, but after that stage the irregularities become more pronounced and the development may take several different directions.

As already mentioned, difficulties are often noted in the separation of the chiasmata at the first anaphase, and consequently chromatin bridges are common. In some cases bridges and fragments indicate that crossing-over has occurred within inverted segments (Fig. 4 *k*). Such configurations were also seen already at metaphase (Fig. 4 *d*, *h* 8). But in the great majority of cases only bridges are present and no fragments. Thus, in 443 b 53 single or double bridges without fragments were counted in 173 cells. And as the frequency of chiasmata in that fixation was 0,65 per cell, obviously more than half of the chiasmata resulted in bridge formation. Also in the other cases where the first anaphase could be studied a varying frequency of bridges without fragments was seen.

While one was formerly inclined to regard these difficulties of separation, which are frequently present especially at first anaphase, as merely accidental stickings, without any far-reaching significance, the studies of McCLINTOCK (1941) and DARLINGTON and UPCOTT (1941) on chromatid breaks and reunions have demonstrated that these dicentric chromatid bridges without fragments may often be interpreted as fusions of broken ends. »Bridges without fragments in cells at meiosis in *Tradescantia*, etc. . . . may be due to sister reunion of true ends rather than to inversion crossing-over» (DARLINGTON and UPCOTT, l. c. p. 334) and »the only occasion in later life when two broken ends (viz. of different chromosomes) are likely to find themselves in contact is at pachytene when, as we shall see, evidence of non-sister reunion of ends is to be found in bridges without fragments at meiosis» (ibid. p. 328). As no bridges are found at the mitoses in haploid rye, it is most likely that non-sister chromatids fuse during the pairing. Thus, it is possible that some of the terminal chiasmata observed at metaphase I actually constitute chromatid reunions. The heterochromatic character of several of the chromosome ends makes it likely that they may be broken during mitotic divisions without severe damage to the

viability. And just the heterochromatic ends are seen frequently to come together at zygotene.

Misdivision of the centromere has been observed rather often at anaphase I. Sometimes a dicentric chromatid has been broken at one of the centromeres (Fig. 4 *m*). This may give rise to one telocentric chromosome at the second division. Misdivision has also been suspected in univalents. It can be established, however, only in the asymmetric chromosomes. But the occurrence of two telocentric chromosomes at second metaphase in one dyad is an indirect proof of this process. Misdivision at the first division of a haploid has been previously observed by HÅKANSSON (l. c. p. 419) in *Godetia*.

IV. THE SECOND DIVISION.

The second division is full of irregularities. In the same pollen sac a variety of stages from the first anaphase to the pollen grains may be found, and the cells lying in the centre of the pollen sac have

TABLE 2. *Chromosome configurations at metaphase II of haploid Secale.*

Configuration No.		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19
Dyad cell 1	Double chromatids	7	6	5	4	5	5	5	5	4	4	4	3	3	3	2	6*	5*	4	5*
	Single chromatids	0	0	0	0	2	2	1	0	3	2	1	2	1	1	3	0	0	1	1
Dyad cell 2	Double chromatids	0	1	2	3	1	0	1	1	1	1	2	2	3	2	2	2*	3*	3*	2
	Single chromatids	0	0	0	0	0	2	1	2	1	2	1	2	1	3	3	0	0	0	0
Number of elements		7	7	7	7	8	9	8	8	9	9	8	9	8	9	10	8	8	8	8
Number of cases		1	9	6	18	1	1	1	2	1	2	6	1	5	2	1	2	1	1	1

often stopped their development already at a prophase stage, usually pachytene.

In some of the fixations the second metaphase could be studied fairly well. In one slide 60 such pollen mother dyads were analysed. The result is recorded in Table 2, and 4 instances of second metaphase are pictured in Fig. 5 *a—d*. From this table and these pictures it will be seen that the second metaphases contain chromosomes with both

double and single chromatids. These types correspond to the chromosomes, which have passed undivided to the poles during the first division, or to those which have been divided. Moreover, a »new» type of chromosomes also occurs, viz. telocentric chromosomes. As already mentioned, they have originated by misdivision. It will be noted that all the centromeres auto-orientate themselves, whether they belong to double, single or misdivided chromosomes. 34 of the 62 cells are found in the 4 first columns of Table 2. In these dyads only chromosomes with double chromatids occur. As might be expected, the distribution 3—4 is most common. It is of interest that even the

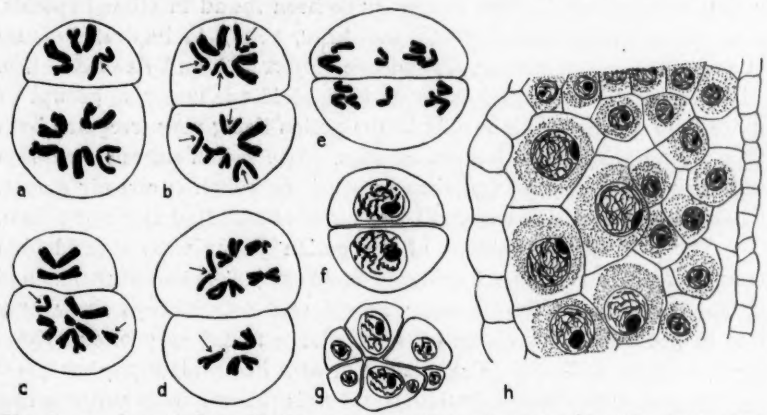

Fig. 5. *a—d*: metaphase II, the arrows indicate chromosomes with single chromatids or telocentric chromosomes, *e*: anaphase II, *f*, *g*: pollen »tetrads», *h*: an instance of chromosome doubling in the sporogenic tissue. — *a—g*: × 1500, *h*: × 600.

distribution 7—0 may give rise to real dyads, one cell of which does not contain any chromatin. In other cases true restitution nuclei are formed, and then the second division is unicellular.

The 23 cells of the columns 5—15 contain different combinations of double and single chromatids. The number of elements in each cell varies from 8 to 10. The only cell with 10 elements is pictured in Fig. 5 *b*. 5 cells with one or two telocentric chromosomes occurred among the recorded material. (Table 2, columns 16—19.) The asterisks in the table indicate that one of the chromosomes was telocentric. The telocentric chromosomes immediately developed a diminutive proximal portion (Fig. 5 *c*, *d*). It is of a certain interest that the telocentric rye chromosome studied by KATTERMANN (1939) also had a small satellite-like »Anhängsel» on the other side of the centromere.

The same thing is true also of the telocentric chromosomes of *Nothoscordum fragrans* (LEVAN and EMSWELLER, 1938).

At the second anaphase the double chromosomes divide and proceed to the poles, while the single chromosomes remain at the equator and later on form micronuclei. Also at the second anaphase dicentric bridges are common, usually without fragments. The result of the second division is very irregular. The tetrads generally consist of 4 cells, it is true, but the cells are very different in size and the larger cells often contain more than one nucleus. Very large monads were sometimes found. Their origin is probably due to fusions of several pollen mother cells. Such fusions have been found in other haploids, e. g. in haploid wheat (GAINES and AASE, 1926), in haploid *Phleum* (LEVAN, 1941) and also in diploid rye (MÜNTZING and PRAKKEN, l. c. Figs. 35—45). Pollen dyads occur in haploid rye and also groups of more than 4 cells. This results in the pollen being very irregular both in size and shape, and it soon atrofies. The pollen mitosis was seen but very rarely, and at that time most of the pollen is already empty. The anthers never dehisce, and I was never able to find any ripe pollen.

The nucleolar conditions of the pollen grains were studied in a couple of slides, stained according to BHADURI (1938) and SEMMENS and BHADURI (1939). While young somatic root cells always show only one nucleolus, older somatic cells with large nuclei very often exhibit several nucleoli (5—8). The nuclear growth has evidently taken place by repeated chromosome doublings, for s_1 is the only chromosome that somatically forms a nucleolus. In the pollen tetrads, on the other hand, I regularly found one nucleolus in each nucleus, even if the nuclei of the tetrad were as many as seven (Fig. 5 *g*). And since s_1 can occur only in 2 of the nuclei of each tetrad it is clear that also other chromosomes than s_1 may develop nucleoli, if they are isolated in nuclei in which no s_1 is present. This is in agreement with what takes place in the abnormal telophases occurring after colchicine treatment. Each nucleus will develop at least one nucleolus.

V. CHROMOSOME DOUBLINGS WITHIN THE HAPLOID.

As already mentioned, the number of the nucleoli indicates that the specialised older tissues of the haploid are often polyploid. In the solitary mitoses found in such tissues the chromosome number 14 and 28 could occasionally be counted, but higher numbers were not found. Evidently cells with higher chromosome numbers do not often divide.

This property of developing polyploid specialised tissues is nothing characteristic of the haploid, but seems to be a common feature in all organisms, plants as well as animals. The occurrence of entire tissues with increased chromosome number was already established by WINKLER in his classical paper of 1916. Recently GEITLER and his collaborators (e. g. GEITLER, 1940 b) have studied this question and also proved that the chromosome doubling takes place by endo-mitoses.

More accidental chromosome doublings were also seen in the studied rye haploids. Thus, the whole nucellus was diploid in one ovule of 317—17. The rest of the flower was normally haploid. Of special interest are those doublings which concern the sporogenic tissue. Such doublings were met with twice. In both cases the doublings had changed several pollen mother cells within one pollen sac.

In the first case two large groups of giant cells were found, one in the middle of the pollen sac, the other at its upper end. Both the normal and the giant pollen mother cells had just began early prophase. Judging from the size difference (Fig. 5 *h*), it is not excluded that the large cells had undergone two doublings and consequently were tetraploid. The difference in nucleolar size was considerable too.

The second case of doubling within the pollen mother cells could be better analysed, because the cells in question had here reached metaphase I. In this case, too, the doubled cells were lying as two separate isles within one pollen sac. If the longitudinal section of this pollen sac is coarsely divided into fourths, the pollen mother cells had the following sequence starting from the lower part of the pollen sac: (1) haploid diakinesis changing upwards into haploid metaphase I, (2) diploid metaphase I, the chromosomes supercontracted, (3) haploid metaphase I, chromosomes of somatic appearance, very little contraction, (4) diploid metaphase I towards the top of the pollen sac passing over into anaphase—telophase I. It is of interest to observe that especially section 3, the haploid isle, enclosed on both sides in diploid tissue, had very abnormal chromosome conditions. The chromosomes were long, despiralised, and of the type found after colchicine treatment of meiotic prophase (LEVAN, 1939, Fig. 1 *d*—*l*). The balance within the tissue has evidently been seriously disturbed, the haploid cells responding more strongly than the diploid cells, although even in the diploid cells the chromosome appearance was hardly normal.

In these doubled cells each of the basic chromosomes occurs twice, and the homozygosity must be even more certain than in any inbred rye line. Judging from the number of doubled cells, the doubling

occurred about 5 cell generations earlier. It is of interest to study the bivalent formation in these doubled cells and compare their chiasma conditions with known inbred lines. I was able to analyse 25 cells. The following number of chiasmata per cell were found:

Number of xata per cell	4	5	6	7	8	9	10	11	12	13	14	15	Total	Mean
Total	—	—	—	—	2	3	4	6	5	2	1	2	279	11,16
Terminal	2	—	5	4	5	3	4	1	1	—	—	—	196	7,84

Terminalisation coefficient: 0,7025.

The following number of univalents and different types of bivalents occurred in the 25 cells:

Univalents: 8
Bivalents with 1 xa: 60
» » 2 xata: 102
» » 3 » : 5
xata per bivalent: 1,5943

The commonest bivalent type was the ring with 2 xata.

LAMM (1936) has found that inbred lines of rye have on an average a lower chiasma frequency than population plants. The mean values of his extensive material amounted to:

		xa frequency per cell	xa frequency per bivalent
Population plants	In the field	15,46	2,21
	In the greenhouse	17,39	2,48
Inbred lines.....	In the field	11,49	1,64
	In the greenhouse	15,02	2,15

Thus, the values of my plant (11,16 and 1,59) are at the lower limit of the values of LAMM's inbred lines cultivated in the field. It is known that the ability of causing a decrease in chiasma number is common to various treatments of meiosis which bring about abnormal physiological conditions of the pollen mother cells. The low number of chiasmata formed in the doubled sectors of haploid rye may be ascribed to the general weakness of the haploid plant.

VI. HOMOLOGY, PAIRING, AND CHIASMA FORMATION.

In studying haploid meiosis we are necessarily brought into contact with one of the most important cyto-genetical problems, viz. the question of the relation of chromosome pairing and chiasma formation to homology. According to the classical opinion, the chromosomes pair during zygotene, because their smallest parts are homologous and therefore attract each other. This can almost be said to be the most important axiom of cyto-genetics. On the basis of this assumption the pairing conditions of hybrids and other unbalanced forms are interpreted: if pairing is established, it is considered to be a proof of the presence of homology. The pairing conditions of the salivary gland chromosomes are interpreted in the same way, the least defectiveness in the pairing is taken as a demonstration of interrupted homology.

Owing to the discovery of non-homologous pairing by McCLINTOCK the classical view had to be somewhat modified. It was then supposed that in general only homologous chromosomes pair, but the tendency of the chromosomes to exist in pairs during meiotic prophase is so strong that, lacking homologous partners, also non-homologous chromosomes may exercise an attraction on each other. It is generally assumed, however, that non-homologous pairing is of a less intimate character than homologous pairing. Thus, it should not be able to give rise to chiasmata but only to spiralisation pairing.

In the monohaploids the absence of homologous pairing is encountered in its most marked condition, every chromosome lacking homologous partner. Therefore the haploids should furnish decisive evidence of what can be achieved by non-homologous pairing. The few workers who have studied pachytene of monohaploids have usually observed double structures. RANDOLPH (1932) in haploid maize interprets the double leptotene threads as longitudinally divided univalents, while McCLINTOCK (1933) states that »much of the doubleness of chromosomes in pachytene must be due to the association of non-homologous parts of chromosomes either within a chromosome (fold-backs) or between chromosomes« (l. c. p. 221). CATCHESIDE (1932) in his *Oenothera* haploid has not been able to make a very close study of early prophase, owing to the unfavourable condition of these stages in *Oenothera*. He writes, however: »The doubleness of the pachytene thread usually can be seen especially at a somewhat late stage. Whether these double portions always represent sister chromatids is somewhat uncertain; possibly some of the double threads seen in the tangle are

actually paired segments of two chromosomes» (l. c. p. 76). BLEIER (1933) in his haploid *Oenotheras* could find no evidence of prophase pairing: »Es kommt Synapsis, aber in der Regel keine Syndese vor» (l. c. p. 133).

Prophase of haploid *Triticum monococcum* was studied by KIHARA and KATAYAMA (1933) and KATAYAMA (1935 a). They did not observe any zygotene pairing, but at diakinesis the chromosomes seemed to be attached end-to-end into different complexes, consisting of 2—7 chromosomes. The writers assume that this pairing has nothing to do with homology. Some of their diakinesis pictures show a striking resemblance to the conditions of haploid rye. I am inclined to regard the end-to-end attachments of the *Triticum* chromosomes as remainders of an earlier spiralisation pairing, thus, in fact, the same thing as the residue pairing of rye. Since the chromosomes of *Triticum* often form greater complexes than those of rye, the detailed course of the zygotene pairing may proceed along somewhat different lines. According to KOSTOFF (1938 b, c), *Triticum monococcum* has heterochromatic segments at both ends of all the chromosomes and this may explain the difference in behaviour between *Triticum* and *Secale*. KOSTOFF writes: »It is most probable then that the end-to-end chromosome conjugation in the haploid *Triticum monococcum* is due to a conjugation between heterochromatic segments of the distal regions» (1938 b, p. 18). Also in haploid *Oryza* (MORINAGA and FUKUSHIMA, 1934) the diakinesis pictures (l. c. Figs. 8—11) undoubtedly show residue pairing.

The first haploid to permit a close study of its prophase stages was *Antirrhinum*, in which ERNST (1939) found a typical pachytene stage with a rather complete chromosome pairing in certain cases. In exceptional cases, for instance, in Tafel VII, Abb. 8, the 8 chromosomes can even be seen to form 4 pachytene bivalents. Ordinarily there occurred, however, numerous irregularities of the pairing. *Secale* is another instance of a haploid with a pronounced pachytene stage. Although I will not assert that this stage must always reach its most advanced condition in haploid rye — it probably very often happens that the pairing is suppressed by certain genotypes and by certain environments — nevertheless, if typically developed, the pairing can acquire almost completeness. Especially suggestive is the remarkable regularity of the course of the pairing found in *Secale*: it starts at the chromosome ends and then proceeds inwards. It may be noted in this connexion that LAMM (1936), because of the distribution of chiasmata in diploid *Secale*, assumes that the pairing is initiated at the

ends also in this form (l. c. p. 229). *These observations on the haploid pachytene pairing make it absolutely indisputable that non-homologous pairing plays a dominating rôle.* The mechanism of non-homologous pairing has been discussed recently by RIBBANDS (1941). Although he admits that the heterochromatic regions may effect an attraction on each other and cause non-homologous pairing in *Zea* and in salivary gland chromosomes, in his own material, meiosis of male *Habropogon* (Diptera), a special mutual attraction existed between chromosome ends and centromeres without any indications of the presence of heterochromatin in these regions.

The next question will be whether this non-homologous pairing can cause the formation of real chiasmata, or if the chiasmata seen in haploids, are all formed within the duplicated segments, which may be supposed to occur within all genomes. It can be said already now, that even if this latter condition is most probable in some haploids, the regular type of pairing in *Secale* undoubtedly constitutes a not insignificant difficulty for this presumption. As already described, it can be directly seen that the bivalents in haploid rye are usually symmetric; this is the case both with the bivalents, held together only by the residue pairing, and those with real chiasmata. And although bivalents with asymmetric chiasmata do occur in rye, as in haploid *Oenothera*, the asymmetry is never very pronounced. Metaphase bivalents in haploid *Triticum monococcum*, as well as in haploid *Hordeum distichum* (TOMETORP, 1939), are of the same type as the fully terminalised rye bivalents. Nor are signs of pronounced asymmetry met with in haploid *Oryza*, *Pharbitis* (KATAYAMA, 1935 b), *Godetia* or any of the known bivalent-forming haploids. One should no doubt have expected a greater irregularity in the appearance of the bivalents, if chiasmata could be formed only within homologous segments, scattered over the genome. If these conditions are to be explained according to the homology hypothesis, rather specialised accessory hypotheses must be resorted to, as, for instance, the location of duplications at symmetric parts of the chromosomes. If one assumes, as CATCHESIDE does in his *Oenothera* haploid, that the chiasmata at metaphase I remain at about the same position as where they were formed, then it becomes necessary to presume a crowding of duplicated segments towards the ends of the chromosomes.

If, on the other hand, the admission is made that also non-homologous pairing can give rise to chiasmata, it may be noted that the genotype of the species in haploid condition reacts in the same

manner as in diploid condition. The pairing starts at the same place, chiasmata are formed in the same region, and the bivalents assume the same type as in the diploid. It is only the number of chiasmata that differs, and the very mechanism of chiasma formation, which is often completely suppressed. There is no difficulty in imagining that non-homologous pairing will be even more easily disturbed than homologous pairing.

It has been shown, among others, by HALDANE (1931) that the chiasma distribution in normal diploid forms does not directly follow a POISSON distribution, as should have been expected considering that each cell contains the same paired chromosome length, along which an indefinite number of chiasmata are to be distributed at random. The deviation from POISSON is caused by the cytological interference, i. e. the chiasmata are not formed independently of each other. The distribution is decidedly excessive, the values being drawn together around the mean. In a haploid organism with its low chiasma frequency very seldom more than one chiasma per bivalent is formed. With the same supposition of a potential number of chiasmata distribution at random over the chromosome length, a POISSON distribution should therefore be valid. Any deviation from POISSON should have a definite cause, another factor than pure chance having interfered.

Below the chiasma formation is analysed in those few monohaploids where suitable material is available. An examination will be made, where possible, to learn the extent to which the found chiasma distributions agree with POISSON. In addition to *Secale*, such data are available in 2 genera, viz. *Oenothera* (CATCHESIDE, 1932; BLEIER, 1933) and *Antirrhinum* (ERNST, 1939).

In Table 3 a survey is given of the different metaphase I configurations in the three *Oenothera* haploids examined. *Oenothera blandina* has 6 different configurations of uni-, bi- and multivalents. The chiasma frequency is 0,24 per cell, thus being within the variation area of *Secale*. CATCHESIDE distinguishes between 4 types of chiasmata at metaphase I, depending on their degree of terminalisation and their appearance. From this he concludes that 4 types of duplications are involved in the chiasma formation. Certain of these segments are assumed to be present more than in duplicates, and certain of the chiasma types may be caused by more than one pair of homologous segments. Still, CATCHESIDE explains the chiasmata by assuming a small number (less than 10) of duplications, *inter se* different in size.

The haploids of *Oenothera Hookeri* and *franciscana*, studied by

TABLE 3. *Metaphase I configurations of haploid Oenothera.*

TABLE 3. *Metaphase I configurations of haploid Oenothera.*

Configuration No.		1	2	3	4	5	6	Total of cells	Total xata	xata/cell
Number of	I	7	5	3	4	2	3			
	II	—	1	2	—	1	—			
	III	—	—	—	1	1	—			
	IV	—	—	—	—	—	1			
	xata	0	1	2	2	3	3			
Oenothera blandina (CATCHESIDE, 1932)	Number of cells	938	219	17	11	1	1	1187	281	0,24
	%	79,0	18,5	1,4	0,9	0,1	0,1			
Oe. Hookeri (BLEIER, 1933)	Number of cells	1121	145	9	—	—	—	1275	163	0,13
	%	87,9	11,4	0,7						
Oe. franciscana (BLEIER, 1933)	Number of cells	2187	143	3	—	—	—	2333	149	0,06
	%	93,7	6,1	0,1						

TABLE 4. *Metaphase I configurations of haploid Antirrhinum.*

Configuration No.		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	Total of cells	Total of xata	xata/cell
Number of	I	8	6	4	2	—	5	3	1	2	4	2	—	1	3	1	2	—			
	II	—	1	2	3	4	—	1	2	—	—	1	2	—	—	1	—	1			
	III	—	—	—	—	—	1	1	1	2	—	—	—	1	—	—	—	—			
	IV	—	—	—	—	—	—	—	—	—	1	1	1	1	—	—	—	—			
	V	—	—	—	—	—	—	—	—	—	—	—	—	—	1	1	—	—			
	VI	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	1	1			
	xata	0	1	2	3	4	2	3	4	4	3	4	5	5	4	5	5	6			
»Diakinesis« (= Metaphase I)	Number of cells	27	87	61	24	—	13	23	—	2	4	5	1	—	2	—	—	1	250	435	1,74
	%	10,8	34,8	24,4	9,6	—	5,2	9,2	—	0,8	1,6	2,0	0,4	—	0,8	—	—	0,4			
Anaphase I	Number of cells	51	100	46	11	2	13	14	1	—	4	3	—	1	1	1	2	—	250	353	1,41
	%	20,4	40,0	18,4	4,4	0,8	5,2	5,6	0,4	—	1,6	1,2	—	0,4	0,4	0,4	0,8	—			

BLEIER, exhibit only three configurations of univalents and bivalents. The chiasma number per cell is lower than in *Oenothera blandina* (0,13 and 0,06 respectively). BLEIER prefers not to draw any conclusions concerning chromosome homology, instead he criticises CATCHESIDE's results in this respect. In BLEIER's opinion, nothing is known about the relation of pairing to chromosome homology. Without knowing the results of MCCLINTOCK, published somewhat later in the same year, he writes as follows: »Z. B. könnte die Paarungsbereitschaft der Chromosomen in der Prophase bei dem Fehlen homologer Chromosomen die Ursache sein, dass bei günstiger Lage zwischen Chromosomen, die zwar nicht, auch in Teilen nicht, homolog sind aber doch eine gewisse chemische Aehnlichkeit besitzen, Paarung stattfinden kann» (l. c. p. 166).

In metaphase of haploid *Antirrhinum* there occurs a variety of no less than 17 different combinations of uni-, bi- and multivalents. In one pictured cell even all the 8 chromosomes are associated into an octovalent. It must be mentioned, however, that the appearance of the septemvalent and octovalent of Fig. 1 *n* and *o* is hardly typical, a chain or a ring configuration would sooner have been expected. The number of chiasmata per cell is high, 1,7 at metaphase and 1,4 at beginning anaphase, thus twice that found in the *Secale* plants richest in chiasmata. ERNST gives a detailed discussion of the question of chromosome pairing and homology. The possibility of *Antirrhinum* being a cryptotetraploid species on the basic number 4 is considered, the haploid then being a pseudo-haploid or allo-haploid. These possibilities are rejected, however, and the author comes to the conclusion that the *Antirrhinum* haploid is a secondary haploid, one or two chromosomes of which are present in double dose, of course more or less changed.

Table 5 shows the chiasma number per cell whithin the haploids analysed. These values are correct in their main features, but the possible presence in solitary cases of more than the necessary minimum number of chiasmata required to form the configurations in question was not taken into account. In my own material four chiasmata in univalents have been disregarded. The values expected according to POISSON are given below each observed series.

It is necessary only to glance at the χ^2 column in order to find that the different haploids behave differently. In the three *Oenothera* materials an almost complete agreement exists between the found and the expected values. The same thing is true of the *Secale* plant with low chiasma frequency. In *Antirrhinum* the deviation from POISSON

TABLE 5. POISSON *test of chiasma frequencies in haploids.*

TABLE 5. POISSON *test of chiasma frequencies in haploids.*

Species			Number of xata per cell							Total of cells	Total of xata	xata/cell	χ^2
			0	1	2	3	4	5	6				
Oenothera blandina		found	938	219	28	2	—	—	—	1187	281	0,2367	
		exp.	936,8	221,7	26,2	2,3	—	—	—				0,1134
Oenothera Hookeri		found	1121	145	9	—	—	—	—	1275	163	0,1278	
		exp.	1122,0	143,4	9,6	—	—	—	—				0,0563
Oenothera franciscana ...		found	2187	143	3	—	—	—	—	2333	149	0,0639	
		exp.	2188,6	139,8	4,6	—	—	—	—				0,6310
Antirrhinum	Metaph. I	found	27	87	74	51	9	1	1	250	435	1,7400	
		$a = 0{,}00$ exp.	43,9	76,4	66,4	38,5	16,8	5,8	2,3				21,1201
		$a = 0{,}70$ exp.	26,5	89,4	78,6	38,4	12,9	3,3	0,8				6,5951
	Anaph. I	found	51	100	59	29	7	4	—	250	353	1,4120	
		$a = 0{,}00$ exp.	60,9	86,0	60,7	28,6	10,1	2,9	0,8				4,5098
		$a = 0{,}50$ exp.	50,2	96,0	66,7	27,2	7,8	1,8	0,3				1,3097
Secale	Many xata	found	557	552	125	14	4	—	—	1252	860	0,6869	
		$a = 0{,}00$ exp.	629,9	432,7	148,6	34,1	5,9	0,8	—				58,0132
		$a = 0{,}40$ exp.	563,9	537,6	131,0	17,7	1,8	—	—				0,8603
	Few xata	found	1189	110	4	1	—	—	—	1304	121	0,0928	
		exp.	1188,5	110,3	5,1	0,1	—	—	—				0,0087

TABLE 6. *Comparison between* POISSON *and binomial distribution of chiasmata.*

No.	Assumption	xata/cell							χ^2 n = 1000
		0	1	2	3	4	5	6	
1	POISSON, total xa frequency = 0,25	0,7788	0,1947	0,0243	0,0020	0,0001	—	—	—
2	Binom. 5 xa points p = 0,05.....	0,7738	0,2036	0,0214	0,0011	0,0000	—	—	1,0974
3	» » » » p = 0,03 — 0,07.....	0,7734	0,2044	0,0211	0,0011	0,0000	—	—	1,2926
4	» » » » $p = 4 \times 0{,}03 + 1 \times 0{,}13$	0,7702	0,2104	0,0187	0,0007	0,0000	—	—	3,7933
5	» » » » $p = 4 \times 0{,}01 + 1 \times 0{,}21$	0,7589	0,2324	0,0086	0,0001	0,0000	—	—	42,6478
6	» 25 » » p = 0,01.....	0,7780	0,1965	0,0238	0,0018	0,0000	—	—	0,0423
7	POISSON, total xa frequency = 1,00	0,3679	0,3679	0,1839	0,0613	0,0153	0,0031	0,0005	—
8	Binom. 5 xa points p = 0,20	0,3277	0,4096	0,2048	0,0512	0,0064	0,0003	0,0000	35,5170
9	» 25 » » p = 0,04	0,3604	0,3754	0,1877	0,0600	0,0137	0,0024	0,0003	0,7921

is not catastrophic, considering the relatively low number of analysed cells. In *Secale* with high chiasma frequency, on the other hand, the deviation is so pronounced that the observed series is devoid of any resemblance to POISSON. The entire excess is gathered in class 1, while all other classes have deficits.

We leave for the present those distributions which follow POISSON in order to look somewhat more closely into the deviating distributions, above all the *Secale* case. The great surplus of the class with one chiasma indicates that a chiasma in a certain place of the idiogram is more easily formed than the other chiasmata. From this it can be concluded that the variation may be divided into two systems: *a*, one certain chiasma, easily formed, and *b*, the rest of the chiasmata. Empirical attempts were made to find out how much of the mean bivalent frequency (0,6869) fell within *a* and *b*, so as to secure the best agreement with POISSON. The frequency within *a* was readily limited to lie between 0,3 and 0,5, and the best result was obtained with a chiasma frequency of 0,4 in *a* and consequently 0,2369 in *b*. As seen from Table 5, the value of χ^2 was changed from 58,0 to 0,9 by this process. Thus it must be said that the distribution found is in good agreement with the assumption that about 60 % of the chiasmata consist of one determined chiasma, which for one reason or other is formed especially often, while 40 % of the chiasmata originate purely at random.

Already the evident resemblance of the deviations from POISSON between *Secale* and *Antirrhinum* (both had a tendency to excessive distribution) indicates that the same method as used in *Secale* should be able to improve the agreement with POISSON of the *Antirrhinum* values. The table shows that this is the case. At both metaphase and anaphase it may be assumed that about 40 % (40,0 and 35,5 % respectively) of the pairing are caused by special chiasmata, while the rest of the pairing is at random. Hereby the χ^2 values decreased from 21 and 5 to 7 and 1 respectively.

Thus, by this analysis it has been established that the three plant genera studied behave differently. While all chiasmata of *Oenothera* follow POISSON, only a certain percentage of the chiasmata of *Secale* and *Antirrhinum* does so. In both these latter genera a heterogeneity of the chiasma formation has been demonstrated.

The next question is: can any conclusions be drawn concerning the cytological nature of the chiasmata from these results? It is evident that a non-homologous pairing, functioning purely mechanically without

regard to chromosome homologies, should be especially predisposed to form random chiasmata. This aspect of the problem does not need any further consideration. Instead we put the question: can also normal homologous pairing bring about a POISSON distribution of the chiasmata?

We consider the simplest case first, viz. *Oenothera*, and test the conditions under which a few fixed chiasma points may cause a POISSON distribution. Suppose that five pairs of duplicated segments occur, each with a chiasma tendency of 0,05. Together they will give a mean chiasma frequency of 0,25 per cell (in CATCHESIDE's material the corresponding number was 0,24). The distribution is obtained from the binome $(0{,}95 + 0{,}05)^5$. Table 6 shows in the first row the POISSON distribution with $\bar{x} = 0{,}25$ and in the second row the binomial distribution. The agreement between them is so great that with a number of individuals of 1000, no difference could be proved ($\chi^2 = 1{,}0974$). CATCHESIDE's interpretation of the chiasmata requires, however, the presence of homologous segments with a varying tendency to chiasmata (calculated from the frequencies of the 4 types of chiasmata: 54,6 25,2, 13,1 and 7,1 %). We test a binomial distribution of five fixed chiasma points with a tendency of 0,03, 0,04, 0,05, 0,06 and 0,07. The third line of Table 6 gives the result. The agreement with POISSON has become somewhat impaired, χ^2 in 1000 individuals has increased to 1,2926. In the series of CATCHESIDE chiasmata occurred in the t-segments more often than in the three other classes together. In line 4 of the table a distribution is tested, where one chiasma tendency has been made 0,13, while the other four are 0,03. The agreement is still worse, χ^2 is nearly 4. The comparison with CATCHESIDE's absolute numbers will be:

Number of xata	0	1	2	3	Total of cells
Found	938	219	28	2	1187
Expected $(0{,}13 + 4 \times 0{,}03)$	914,2	249,8	22,2	0,8	1187,0

$\chi^2 = 6{,}5476$. Row 5 of Table 6 shows the result if one duplication is entirely dominating. One of the five fixed chiasma points has been made $= 0{,}21$ and each of the other four $= 0{,}01$. Now there is no longer any resemblance to POISSON, and χ^2 is $= 42{,}6478$. In fact, a series has been constructed, which resembles my original *Secale* distribution.

So far the number of chiasma points has been kept constant. How would an increase in number of the points act? If the other factors are unchanged, the agreement with POISSON will be improved. In row 6 of Table 6 it is assumed that the same chiasma frequency of 0,25 is caused by 25 fixed chiasma points, each with the tendency of 0,01. Now an almost exact POISSON distribution is obtained ($\chi^2 = 0{,}0423$). If, on the other hand, the absolute chiasma frequency is increased, other factors being constant, the similarity to POISSON decreases. Rows 7 and 8 of the table illustrate this condition. Here the mean chiasma frequency is made 4 times higher than in the earlier series. With 5 fixed chiasma points with equal effect we obtain a χ^2 of 35,5170. The higher the chiasma number, the easier it is to establish any deviations from POISSON. Probably this condition brings about the difference between the two rye materials with different chiasma frequency. If the number of chiasma points with the higher chiasma frequency is increased, the agreement with POISSON again improves. In the ninth row of the table is shown the result of 25 equal chiasma points with a total chiasma tendency of 1,00. The agreement with POISSON is good ($\chi^2 = 0{,}7921$).

We therefore conclude that in the concrete cases examined the establishment of a POISSON distribution of the chiasmata cannot settle the question, if the chiasmata are caused by non-homologous pairing acting at random or by pairing within homologous segments. But certain indications may be given how these homologous segments must be conditioned if a POISSON distribution is present. Thus, their number must not be too small, their chiasma tendency must be about equal and the higher the total chiasma frequency, the greater the number of homologous segment pairs that must be present.

VII. CONCLUSIONS.

Although the pachytene pairing in monohaploids is undisputably demonstrated so far only in *Antirrhinum* and *Secale,* there is reason to believe that it is a generally occurring phenomenon. This pairing is to its greatest extent non-homologous. In most haploids it has tracelessly disappeared before metaphase, but in haploids with long chromosomes it may remain during the whole prophase as residue pairing. Real chiasmata are formed only in a few of the studied monohaploids, and in a relatively great number only in a couple of genera. It must also be remembered that certain of the remaining »chiasmata»

may in fact be due to chromatid reunions. It has not been decided whether the non-homologous pairing really gives rise to chiasmata, but certain reasons seem to indicate that it does so. That some of the chiasmata in haploids are certainly formed within homologous segments is made clear, as pointed out by DARLINGTON (1937), by the inversion bridges met with in haploid anaphase (*Triticum*, *Hordeum*, *Secale*). Perhaps the situation is such that the very start of the pairing is decisive: if the specific attraction between homologous chromomeres becomes predominating, the pairing will begin in the homologous segments, which then get their chance to form chiasmata; if, on the other hand, the non-specific attraction between, for instance, the heterochromatic knobs prevails, the pairing starts in the heterochromatic regions and continues further along the entire chromosome length. Then there is less chance that homologous segments will meet. In most cases these two forces probably cooperate. If, during the course of the non-homologous pairing, two homologous segments happen to meet in an inverted position, there is evidently a great possibility of their forming an inversion loop.

Therefore the possibility that chiasmata may be formed by non-homologous pairing cannot be entirely disregarded, and from this follows that every caution must be used in interpreting homologies from observed chiasmata at metaphase. Above all, there is no reason to assume that entire chromosomes are duplicated in the genome, only because bivalents are formed in the haploid. In *Antirrhinum* and *Secale*, where the chiasma frequencies indicated that one chiasma was more easily formed than the rest of the chiasmata, there are of course reasons to suspect the presence of one or two large duplications as the cause of the heterogeneity of the chiasma distribution. In *Secale*, however, the other fact must also be considered, which should be able to alter the chiasma distribution in just the same manner as one large duplication, namely, the development of dicentric chromatids without chiasma formation. The possibility that the bivalents of *Secale* are of two kinds, normal bivalents with chiasmata and pseudo-bivalents held together by fusion of chromatids, must be reckoned with.

The problem of the original basic chromosome numbers in different plant groups has often been taken up to discussion during recent years. These speculations are sometimes based on the occurrence of polymeric factors, sometimes on the finding of amphibivalent formation at meiosis, sometimes on the discovery of two chromsomes with similar chromosome morphology within the genome, etc. Thus, FLOVIK

(1938) summarises the different reasons why all the Gramineæ should be derived from original types with the basic number 5, and PATHAK (1940) arrives at the same result concerning *Secale* from purely chromosome morphological reasons. It must be said, however, that *a priori* this mechanism involving the doubling of whole chromosomes is too coarse. Especially with such a low number as 5 the trisomic and still more the tetrasomic must acquire an extremely poor vitality, and immense difficulties must be met with before the new form may obtain a balance with its higher chromosome number. The assumption of numerous small changes seems to be decidedly more probable, such as inversions and duplications, which are known for certain to be everyday happenings during the evolution of *Drosophila*. Each step of such a development may be adjusted in relation to the rest of the genotype before the next step is taken. And these processes may well involve changes of the chromosome number, as new centromeres may originate, for instance, by misdivision. It is nevertheless certain that these small changes can hardly be expected to leave such schematic traces in the idiogram, that simply by determining the gross chromosome constants, the position of the centromeres, the number of satellites, the number of secondary constrictions and so on, it will be possible to reconstruct the course of the changes, especially as related groups, sections and whole genera often have genetically fixed a decided uniformity of idiograms, outside of which no changes can occur without resulting in non-stability.

SUMMARY.

1. The heterochromatic regions are shown by cold treatment to be terminally located in the shorter arms of 5 of the chromosomes of the rye genome (Chapter I).

2. A typical pachytene is found in meiotic prophase of haploid rye. The pairing often seems to start at the heterochromatic end knobs. It is mainly non-homologous pairing.

3. At diplotene and diakinesis bivalents and trivalents very often are present. They are held together by the relational spiral (residue pairing) and by chiasmata. The bivalents are as a rule symmetrical, both ends being at the same level. The residue pairing gradually disappears during the progress of prophase and only the pairing by chiasmata remains into metaphase I (Chapter II).

4. The chiasma frequency at metaphase varies from plant to

plant (from 0,08—0,83 chiasmata per cell; Table 1), and within the same plant. Univalents, bivalents, trivalents and quadrivalents are recorded in ten different combinations.

5. At first anaphase chromatin bridges without fragments are often seen. Their possible origin by chromatid reunion is discussed (Chapter III).

6. The second division is usually 2-cellular and often irregular (Table 2). The occurrence of telocentric chromosomes indicates that misdivision of centromeres has taken place at first anaphase (Chapter IV).

7. Sectors with double chromosome number were sometimes found in the haploids. In two cases the doubling had occurred within the sporogenic tissue. The chiasma formation in a doubled sector was compared with the chiasma conditions of inbred rye (Chapter V).

8. An analysis is made of the chiasma conditions of earlier known haploids with chiasma formation (Tables 3 and 4). It is shown that in some cases all the chiasmata, and in other cases some of them, obey the POISSON distribution (Table 5). The bearing of this on the interpretation of the cytological basis of chiasma formation is discussed.

9. It cannot be decided whether non-homologous pairing can give rise to chiasmata. It is shown, however, that certain conditions must be fulfilled, if chiasmata within duplicated segments are to be able to give POISSON distribution (Table 6, Chapter VI).

10. The hypothesis, put forward earlier, that *Secale* is a secondary diploid with an original chromosome number 5 is criticised. An evolution involving numerous small changes seems to be more likely (Chapter VII).

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THE CHROMOSOMES OF TWO SPECIES OF ANABANTIDÆ (TELEOSTEI), WITH A NEW CASE OF SEX REVERSAL

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THE material for this investigation consisted of testes of *Betta splendens* REGAN and *Macropodus opercularis* L. The animals were decapitated, the abdominal cavity opened, the testes removed, placed in the fixative and immediately cut into small pieces. As fixative we used FRIEDMAN and GORDON's modification of NAVASHIN's fluid. The embedding was made in paraffin over methylbenzoate and the slices cut at 10 μ. The stain used was EHRLICH's haematoxylin.

We also used permanent smear preparations of young gastrula stages of *Betta*, made as a modification of BELLING's azeto-carmine method (SVÄRDSON, 1941).

The optics used was a Zeiss' binocular microscope with a Leitz' apocr. obj. $\times$ 112, apert. 1,32 with Zeiss' compens. ocul. $\times$ 20.

Mitosis. — The spermatogonial mitosis completely follows the normal scheme. Owing to lack of material we were unable to determine the chromosome number in the spermatogonia of any of the species. The embryos of *Bettà*, however, gave some figures, showing that the number of chromosomes must be $2n = 42$, the same number as that found by BENNINGTON (1936). The chromosomes of the embryonic mitoses of *Betta* are short, the largest (at metaphase) being about 3 μ. Most of the centromeres seem to be terminal, but at least 3 pairs of chromosomes are two-armed and therefore must have subterminal centromeres. BENNINGTON was unable to determine the morphology of any chromosome, and in his figures all of them are drawn as short rods.

Some mitotic figures in a haploid embryo of *Betta* were detected and were found to contain only 21 chromosomes.

Meiosis. — The prophase of meiosis is completely normal. No polarisation is found in the zygotene stage. After pachytene, which as usual is of a very long duration, there follow a diffuse diplotene and a diakinesis. The chromosomes are not stainable and visible until

late diakinesis, when they are distributed in close connection to the nuclear membrane. This is in agreement with the findings in Cyprinodontidæ (WICKBOM, unpubl.).

No pair of chromosomes shows any peculiar behaviour, such as precociousness, etc. during the meiotic prophase.

At metaphase the bivalents are arranged as usual with the large ones in the periphery and the smaller ones in the centre. The two-armed structure of some chromosomes is very evident in side views of the metaphase, where these chromosomes are hold together by chiasmata in their long arms, while the short arms are free.

The number of bivalents of the two species investigated is 21. As far as *Betta* is concerned this is in agreement with BENNINGTON's report (1936). *Macropodus* has not been investigated before.

In neither of these species did we find any chromosome pair behaving at metaphase and anaphase like a pair of heterochromosomes. This is not in accordance with BENNINGTON's findings, but as this question is dealt with in a previous paper (WICKBOM, 1941) we will not enter into it here.

With regard to the homology of chromosomes between the two species, which are very closely related to each other, it is impossible to say anything for certain at the present standpoint. Good mitotic figures from both species and the meiosis of the hybrid between them must be investigated first. But the number of chromosomes and the similarity of the meiotic stages of the two species indicate that their chromosome sets must be alike. It seems to be a significant feature of lower vertebrates that the chromosome number and the morphology of the chromosomes are alike or identical if the species belong to the same or closely related genera, e. g. *Pungitius* species (MAKINO, 1933), Cyprinodontidæ (FRIEDMAN and GORDON, 1934; RALSTON, 1934; WICKBOM, 1941), *Rana* and *Bufo* (OGUMA and MAKINO, 1937), etc. It is, however, too early to draw any phylogenetical conclusions from these facts, especially as the chromosomes of Vertebrata are very incompletely known.

Sex reversal. — In the testis of one individual of *Betta splendens* we found a great number of residual bodies (Fig. 1), showing that this animal must have some time ago passed through a sex reversal from female to male. Such a sex reversal is known from many Teleosts and Anurans (REGNIER, 1938 et lit. ibidem cit.). The gonad in question was a well-developed, functioning testis with all stages of spermatogenesis.

The animal had previously been used for breeding and the litters contained both sexes, though the females were in the majority. The number of young, however, is too small to draw any conclusions as to the mechanism of the sexual determination of *Betta*. Thus it must be left undecided whether this species is phenotypically sexually determinated like *Xiphophorus helleri* (KOSSWIG, 1935), or if sex reversal of adult individuals are only exceptions such as those described from *Lebistes* and other Teleosts (lit. in REGNIER, 1938). Sex reversal is

Fig. 1. Testis with residual bodies of a *Betta* male. It is evident that this animal must have passed through a female stage. — × 300.

already known from *Betta* through experiments by NOBLE and KUMPF (1937), who found that after extirpation of the ovary a testis regenerated from the oviduct. The progeny after such a »male» mated with a normal female was 9 ♂♂ : 12 ♀♀. SCHMIDT (1930) reports cases of sex reversal from *Betta* and *Macropodus*. Here, too, the females became males but in a natural manner, not induced by experiments. The progeny from a reverted male × a normal female was 32 ♂♂ : 43 ♀♀. SCHMIDT states that the reverted male lacks the instinct for building a nest. This was also true concerning a male of *Macropodus*, which had passed through a female stage. The progeny after this male was

74 ♂♂ : 197 ♀♀. Sex reversal is also known from a third genus of Anabantidæ, *Trichogaster* (SCHMIDT, 1930).

Undoubtedly these very common cases of sex reversal in Teleosts are due partly to the undeveloped sex chromosome mechanism, which is evidently characteristic of the whole Caldus, as for Amphibia, and partly to the embryonic hermaphroditism, which is reported from almost every Teleost investigated with regard to this detail of the gonad development. MRŠIĆ (1931) found it in *Salmo*, DILDINE (1936) in *Lebistes*, BREIDER (1935) in *Limia* species, KOSSWIG (1935) in *Xiphophorus* and *Platypoecilus variatus*, and REGNIER (1938) in *Xiphophorus*. HOFMEISTER's (1939) investigation of the Salmonid *Osmerus* indicates its presence also in this species. WICKBOM (unpubl.) found stages which may indicate that *Alburnus*, too, passes through a protogynous stage.

That the sex chromosome mechanism of *Xiphophorus helleri* must be very undeveloped is claimed by KOSSWIG (1935). The same thing was found in *Limia* by BREIDER (1935). These investigations are purely genetical. The cytological conditions of the sex chromosomes of Cyprinodontidæ, *Betta* and probably of Teleosts in general are discussed by WICKBOM (1941). Many facts indicate that the same peculiarities may explain the conditions of the sex differentiation and sex chromosomes of Anurans (GALGANO, 1933), although they have been the subject of much discussion up to the present time. (Lit. in OGUMA and MAKINO, 1937.)

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SUMMARY.

1. The chromosome number of *Betta splendens* REGAN is $2n = 42$, $n = 21$; the same as reported by BENNINGTON (1936).

2. The chromosomes of *Betta* are short, less than 3 μ, and have terminal centromeres, except 3 pairs, which are two-armed.

3. The chromosome number of *Macropodus opercularis* L. is $n = 21$. Mitosis has not been investigated owing to lack of material.

4. No sex chromosomes have been detected, neither with respect to behaviour nor morphology.

5. A case of sex reversal in *Betta* is described and is associated with previously known cases in Anabantidæ and other Teleosts. The possible connection between the sex reversal, the protogynous hermaphroditism and the undeveloped sex chromosomes is discussed.

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FREQUENCY OF INDUCED CHLOROPHYLL MUTATIONS IN DIPLOID AND TETRAPLOID BARLEY

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SEEDS of a variety of barley (Opal B) and of the corresponding autotetraploid strain were irradiated in 1940. The immediate effects of this treatment have already been described (MÜNTZING, 1941). In the tetraploid material the germination of the seeds and the vigour of the resulting plants were on an average much less decreased than in the corresponding diploids. Also as regards fertility the average decrease was less pronounced in the tetraploids than in the diploids. This differential behaviour was ascribed to the fact that in the autotetraploid line there are four quite homologous genomes instead of only two as in the diploid. Thanks to this gene and chromosome duplication deleterious changes occurring at a certain locus in one chromosome will in most cases be counterbalanced by a normal condition in the other homologous chromosomes.

This view was further supported by observations of the X_2-generation, grown in 1941. In the first place the production of chlorophyll mutations is of interest. When observing the seedlings in the field a distinct difference was at once evident between the diploid and tetraploid material. In the tetraploid plots not a single typical chlorophyll mutant could be detected. In the diploid plots, on the contrary, there was a rather frequent occurrence of the usual categories of chlorophyll mutations, viz. *albina*, *xantha*, *alboviridis*, *viridis* and *tigrina* (cf. GUSTAFSSON, 1940). The data available may be summarised in the following table (Table 1).

As is evident from the table, 75 chlorophyll mutants were present in the X_2-generation of the diploids among a total of 5,558 plants observed. These values were obtained by summation of the figures corresponding to 5,000, 10,000 and 15,000 r. In the tetraploids, 2,365 plants belonged to the X_2-progenies corresponding to the same intensities of irradiation (5,000—15,000 r). All these 2,365 plants were apparently normal, or, at any rate, they did not represent the clear-cut mutant categories occurring in the diploid material. Thus, the com-

TABLE 1. *Chlorophyll mutations in X_2 of diploid and tetraploid barley.*

Category	Number of families	Number of seeds sown	Number of plants obtained	Percentage of plants	Number of families with mutations	Total number of mutants	Percentage of families with mutations
Diploid, untreated ...	—	690	413	59,86	—	—	—
» , 5,000 r	102	3269	1609	49,22	10	18	9,80
» , 10,000 r	104	2974	1697	57,06	20	32	19,23
» , 15,000 r	115	3898	2252	57,77	16	25	13,91
Tetraploid, untreated	—	780	236	30,26	—	—	—
» , 5,000 r	105	2161	849	39,29	—	—	—
» , 10,000 r	102	1670	679	40,66	—	—	—
» , 15,000 r	105	2277	837	36,76	—	—	—
» , 20,000 r	98	2480	521	21,01	—	—	—
» , 25,000 r	112	2136	471	22,05	—	—	—

parison between the diploid and tetraploid material may be concentrated to the following figures:

	Total number of individuals	
	Mutant	Normal
Diploid:	75	5,483
Tetraploid:	0	2,365

A calculation of χ^2 gave the value 32,20 which corresponds to a P much smaller than 0,001. Thus, the difference between the distributions is significant. A still higher χ^2 value would have been obtained, if the tetraploid families corresponding to 20,000 and 25,000 r had been included.

The contrast between diploids and tetraploids may also be given in terms of the percentage of X_2 families segregating chlorophyll mutants. As is evident from the table, these values ranged from 9,80 to 19,23 in the diploids and, as already mentioned, were 0,00 in all tetraploid categories. A calculation of χ^2 in this case (for the dosages 5,000—15,000 r) will be based on the following values:

	Number of families	
	With mutations	Without mutations
Diploid:	46	265
Tetraploid:	0	312

The resulting χ^2 value will be 49,83, P being much less than 0,001. This comparison, however, is less correct than the preceding one, as the number of individuals in the tetraploid families was only about

half that in the diploids. The small number of individuals in the tetraploids is due to the rather pronounced sterility characteristic of this autotetraploid material.

Though no chlorophyll mutations of the striking kind occurring in the diploid material were observed in the tetraploids, some other deviations from normal chlorophyll development in the tetraploids should be mentioned. In a low proportion of plants the leaves were not uniformly green but had irregular yellowish spots or broad transverse zones, not sharply differentiated from the purely green tissue. As plants of this kind also occur in the untreated tetraploids it is probably a characteristic property accompanying the tetraploid condition and not an effect of the X-ray treatment. In the tetraploid material, corresponding to the doses 5,000, 10,000, 15,000, 20,000 and 25,000 r the percentage of such plants was found to be 1,77; 0,74; 0,36; 0,19 and 0,00 respectively. — The apparently regular decrease in this series of values is probably without significance. At any rate, the frequency of such chlorophyll abnormalities is not increased by stronger doses of irradiation. On an average it is not higher than in the untreated tetraploid material, which was found to contain 0,85 per cent of plants of this kind.

The absence of true and visible chlorophyll mutants in the tetraploid material may partly be ascribed to the low average number of individuals per progeny. This number ranged from 8,1 to 4,2 in the tetraploids and from 19,6 to 15,8 in the diploids (cf. Table 1). However, since the total number of tetraploid plants observed was as high as 3,357 there should nevertheless be ample opportunity for the segregation of recessive individuals from duplex heterozygotes, *AAaa*. Thus, as was to be expected, the irradiation in our tetraploid barley only resulted in plants having one of the four homologous chromosomes changed at the same point. In the offspring of such *AAAa* heterozygotes theoretically, it is true, a low proportion of *aaaa* individuals might be formed, but the material now studied evidently was not extensive enough to show the production of such individuals. Nevertheless, this material was quite sufficient to demonstrate the differential reaction of tetraploid and diploid barley to irradiation also as regards the production of chlorophyll mutations.

Though the study of chlorophyll mutations in the diploid material is beyond the main scope of the present investigation a few words may also be said about the mutant categories found and the correlation between fertility of the X_1-plants and category of mutation in X_2. The

mutants were classified according to the key given by GUSTAFSSON (1940, p. 4). The following types and frequencies were observed:

Category	Number of families studied	Number of families segregating				
		albina	*xantha*	*alboviridis*	*viridis*	*tigrina*
diploid, 5,000 r	102	5	—	—	5	—
» , 10,000 r	104	10	4	4	6	1
» , 15,000 r	115	4	5	1	5	1

In four diploid control plots, comprising a total of 413 plants, no mutants occurred.

According to the above values, the *albina* mutations seem to be relatively more frequent at lower dosages than the other mutations, the percentage of *albina* mutations at 5,000, 10,000 and 15,000 r being 50, 40 and 25 respectively. The numbers now observed, however, are too small to give significant differences, but at any rate they point in the same direction as the results obtained by GUSTAFSSON (1940, p. 18) on a more extensive material. According to GUSTAFSSON (l. c.), *albina* mutations do not increase proportionally to the dose, in contrast to the *xantha*, *alboviridis* and *viridis* mutations. This is remarkable, since evidence was obtained that the *albina* mutations represent intragenic changes, whereas *xantha* and *viridis* are associated with or conditioned by structural chromosome changes. This evidence mainly consists in the observation that *xantha* and *viridis*, in contrast to *albina*, only rarely appear in the offspring of quite fertile X_1 plants (GUSTAFSSON, 1940, pp. 8—14). Consequently, the X_1 plants giving rise to *albina* mutations should, on an average, have a better fertility than those producing *xantha* and *viridis* mutations. This was tested in my present material, the following result being obtained:

	Percentage of seed setting in the mother plant																			n	M
	25	—	30	—	35	—	40	—	45	—	50	—	55	—	60	—	65	—	70		
albina	1	—	—	—	1	—	2	2	—	—	4	5	1	3						19	73,81
xantha + *viridis* ...		1	—	2	1	2	2	3	3	2	—	6	2	1						25	72,30

Evidently these series do not reveal any significant difference between the two categories. However, in view of the rather convincing though perhaps not quite conclusive evidence obtained by GUSTAFSSON (l. c.) it is possible that a larger material might reveal a difference of the

kind expected. It should also be observed that data from field experiments with a rather low degree of seed germination (Table 1) give less reliable results than experiments carried out under more favourable conditions. Still the probable correlation between different degrees of sterility in X_1 and different categories of mutation in X_2 must at any rate be rather weak.

According to Table 1 there is a good proportionality between dosage and mutation frequency for the doses 5,000 and 10,000 r, the percentage of families with mutations being 9,80 and 19,23 respectively. As two of the families corresponding to 10,000 r segregated two different mutations each, the percentage of mutations in the X_1 plants treated with 10,000 r was in reality slightly higher than 19,23, the correct value being 21,15. The ratio 21,15 : 9,80 is 2,16. This good proportionality between dosage and mutation frequency is not at all valid for the strongest dose, 15,000 r, the mutation frequency in this case being as low as 13,91 (Table 1). There is another kind of correlation, however, which is valid also for the dosage 15,000 r, viz. the correlation between degree of sterility in X_1 and mutation frequency in X_2. According to Table 1 in my previous paper (MÜNTZING, 1941) the average percentages of seed setting in X_1 for the dosages 5,000, 10,000 and 15,000 r were 77,71; 64,85 and 73,17 respectively. These values evidently show good proportionality to the frequencies of mutations observed in X_2 (9,80; 21,15 and 13,91 respectively). A similar proportionality between degree of sterility and mutation frequency has already been observed by GUSTAFSSON (1940) and GELIN (1941). GELIN finds a very marked parallelism between the percentages of cells in X_1 showing cytological disturbances, the degree of sterility in X_1 and the mutation frequency in X_2. This strongly indicates that most of the induced mutations in barley are associated with or caused by structural chromosome alterations.

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ABSTRACTS — KURZE MITTEILUNGEN

GUNNAR NILSSON-LEISSNER: A case of increased vitality in sib-pollinated later generations of self-fertilised *Dactylis glomerata* strains.

Professor NILS SYLVÉN, my predecessor as head of the herbage plant breeding in Svalöf, always tested seed from the institution's own seed plots of the respective grass strains side by side with seed of the same strains from the General Swedish Seed Company, who carries out all large propagations of varieties from the Swedish Seed Association. In a series of years it was observed in these yield tests that seed of the I_1 generation of strains of *Dactylis glomerata*, originating from self-fertilisation of a single plant, generally produced weaker plant stands than seed of later generations after open pollination in locally isolated seed fields. The differences in the field were also rather striking, as the I_1-plots always had small blanks in the plant rows and plants with more or less curly leaves or with other minor abnormalities were often found in them. The growing rate of the plants also seemed to be somewhat retarded. This was, however, not the case in plots from seed of later generations. The yield figures illustrated the same phenomenon.

In order to investigate the problem conclusively I have now analysed all such available experiments with two *Dactylis glomerata* strains, Skandia II and Brage, both now placed on the market, in which the later seed generations could be followed in every detail. Each of these varieties originates from one single plant, which was at first propagated vegetatively in locally isolated plots in the experimental fields of the institution. Seed from these plots is used partly for yield tests and is partly delivered to the Seed Company for propagation in large, carefully isolated fields. In the first year these seed fields are small, depending upon the amount of seed available from the vegetative propagations, but are soon enlarged by sowing the seed harvested from the first fields close up to them, thus establishing large seed propagations, containing plants of two different generations. For this reason it has unfortunately not been possible to follow the seed generations step by step, but only to compare the I_1-generation with the 3 following generations. — The yield tests in question have been carried on since 1925 with only occasional interruptions. They were drilled in rows 20 cm. apart with a plot size of 10 m^2 and 4 to 5 replicates. Cover crops were barley or early oats. The trials were, with but few exceptions, harvested twice during two consecutive harvest years. In the table only the total yield of green matter in the respective tests are compared. To avoid errors due to the different seed quality of the lots compared, the amount of seed per unit area was compensated according to purity, germinability, and weight of 1000 grains in the manner usually accepted for field trials. In two cases the differences in seed quality were rather great; in spite of the compensation of the amount of seed, these trials were omitted, as it might be suspected that full equality between the seed lots could not be attained in this way. 12 trials with the variety Skandia II and 10 with Brage were left for comparison.

The results concerning quality of seed and yield of green matter are

presented in Table 1. As differences in purity of seed can be fully compensated by adjusting the amount of seed per unit area, such figures are of little interest in this connection and were therefore excluded from the table. It may, however, be stated that in both varieties the mean purity of the I_1-seed is the highest as compared with the seed of later generations. The differences are only small. No significant differences in germination percentage can be recorded, but as regards seed size the I_1-seed was, with but few exceptions, superior in Skandia II and inferior in Brage. It should be pointed out that in the former case, where the I_1-seed is superior, the inferiority in yield of plant stands originated from this seed is greater than in the latter. Besides, compensations in amount of seed were made for these differences. The yield differences for both varieties are to the advantage of later seed generations and in the case of Skandia II fully significant ($P = 0{,}02$). For Brage the mean difference is not much smaller but more variable and therefore no significance can be established ($P = 0{,}1$). As the two series of differences are rather similar and have about the same means, we should be justified in combining them. In that case the mean yield difference is 1,630 kilos per hectare and is very significant ($P = 0{,}01 - - 0{,}001$).

From what has been said above it is quite clear that in the case of Skandia II and Brage, cocksfoot seed of the I_1-generation gives raise to a weaker plant stand than seed of later generations isolated from other cocksfoot strains. There is no reason to ascribe this weakness to differences in seed quality or treatment. From similar experiments (unpublished) in other grass species, e. g. timothy, meadow fescue and red fescue, it is also evident that such an increase in vigour in succeeding generations is very difficult to demonstrate, if it exists at all. No morphological abnormalities were observed in the I_1 of these species. It seems very difficult to find an explanation of the peculiar behaviour of the cocksfoot strains according to commonly accepted theories of the mechanism of loss of vigour by inbreeding, heterosis by intercrossing

TABLE 1. *Means of I_1 and later sib-pollinated generations (I_2—I_4) of Dactylis glomerata.*

	No. of tests	Germination %				Weight of 1000 grains; g				Yield; kg per hectare			
				Diff.				Diff.				Diff.	
		I_1	I_2—I_4	%	D/m	I_1	I_2—I_4	g	D/m	I_1	I_2—I_4	kg	D/m
Skandia II	12	94,08	91,75	+ 2,33	1,6	1,20	1,00	+ 0,11	2,8	31,140	32,960	— 1,820	2,7
Brage	10	93,90	93,60	+ 0,30	0,25	0,89	0,96	— 0,29	3,2	36,710	38,120	— 1,410	1,8
Both varieties	22	—	—	—	—	—	—	—	—	33,670	35,500	— 1,830	3,3

of inbred strains, etc. The following theory of natural selection is therefore put forward.

Evidently some weak, more or less abnormal types segregate by the inbreeding of the *Dactylis glomerata* strains in question. This can be seen as blanks or as plants with abnormal leaves, etc. in the I_1-plots. In a seed field with dense plant rows the weak plants are suppressed and will never reach seed setting. This will be repeated in all subsequent generations, working as a continuous selection for strong and normal types and causing an improvement of the mean vitality of the plants. In the other species mentioned, where the weakness of the I_1-generation was less striking, such a natural selection does not result in demonstrable improvements.

This report may be of some interest to both geneticists and practical plant breeders, as it calls attention to the difficulty of comparing successive inbreeding generations and the necessity of letting families and strains of cross-fertilising plant species adapt themselves to the environment before the final testing.

Svalöf, Swedish Seed Association, November 1941.

FOLKE FAGERLIND: Kommt Agamospermie bei den *canina*-Rosen vor?

Neulich lieferte ich (FAGERLIND, 1940) eine Zusammenstellung der Ergebnisse von Studien über *canina*-Rosen und Versuchen mit solchen, die von verschiedenen Forschern und mehrmals in der Absicht ausgeführt worden sind, das Vorkommen einer agamospermischen Vermehrungsweise nachzuweisen. In dieser meiner Zusammenstellung zeigte ich, dass sichere Beweise für das Vorliegen der genannten Erscheinung in keinem Falle erbracht worden sind. Kreuzungsversuche, die ich ausgeführt habe, und deren vorläufige Ergebnisse in der obengenannten Arbeit mitgeteilt wurden, sprechen stark dafür, dass Mixis vorliegt.

Das Samenmaterial (Nussmaterial), das bei den erwähnten Kreuzungsversuchen erhalten wurde, ist ausgesät worden. Das nun zur Verfügung stehende F_1-Material besteht nur aus einer geringen Anzahl Pflanzen. Es beruht dies teilweise darauf, dass die Keimfähigkeit sehr schlecht war, vor allem aber darauf, dass das Samenmaterial nicht die Vorbehandlung vertragen hatte, der es unterzogen worden war, um die lange Keimzeit zu verkürzen. Der grössere Teil desselben wurde nämlich von der Nussschale durch Abschleifen mittels eines zahnärztlichen Werkzeugs (Karborundrädchen) befreit. Bei einer etwas kleineren Fraktion wurde die Schale partiell weggeschliffen. Nur ein kleiner Teil des Materials wurde ausgesät, ohne dass eine Vorbehandlung erfolgt war. Leider haben die den beiden grossen Fraktionen angehörigen Samen ihre Entwicklungsfähigkeit vollständig eingebüsst. Die kleine, unbehandelt gebliebene Fraktion bestand nur aus Nachkommen der Kombinationen *Rosa rubrifolia* × *rugosa*, *R. rubrifolia* × *pendulina* und *R. villosa* × *rugosa*. Die erstgenannte F_1-Gruppe ist bisher nur durch 8, die mittlere durch 10 und die letzte durch 3 Individuen vertreten.

Die den zwei ersten F_1-Gruppen angehörigen jungen Pflanzen sind ein-

ander augenfällig ähnlich. Sie zeigen auch vollständige Übereinstimmung mit jungen *R. rubrifolia*-Pflanzen. Was den für diese Art so charakteristischen Anthozyangehalt betrifft, so scheint er bei den erwähnten F_1-Pflanzen nicht, und sei es auch nur ein wenig, abgeschwächt zu sein. Bei einer äusserlichen Besichtigung der beiden Gruppen liegt daher der Gedanke an apomiktisch gebildete Nachkommenschaft sehr nahe. Eine zytologische Untersuchung zeigt jedoch, dass eine derartige Schlussfolgerung unzutreffend ist. Die Pflanzen der ersten Gruppe haben, wie *R. rubrifolia*, 28 Chromosomen. Alle Pflanzen der zweiten Gruppe haben 35 Chromosomen. Da die Chromosomenzahl bei *R. rugosa* $2n = 14$ und bei *R. pendulina* $2n = 28$ ist, muss Mixis stattgefunden haben. Die 21 Chromosomen enthaltenden Embryosäcke der *R. rubrifolia* müssen von den 7 bzw. 14 Chromosomen enthaltenden männlichen Kernen der *R. rugosa* bzw. *pendulina* befruchtet worden sein.

Die bei den *canina*-Rosen oft erwähnte Konstanz muss auf dem Vorliegen ausgesprochener Dominanzerscheinungen beruhen. Diese stehen natürlich in Zusammenhang mit den Verhältnissen zwischen den von den Mutterpflanzen und den von den Vaterpflanzen herstammenden Chromatinmengen. Eine Äusserung einer solchen Dominanz ist offenbar der starke Anthozyangehalt bei den obenerwähnten *R. rubrifolia*-Nachkommen. Dozent ÅKE GUSTAFSSON teilte im Anschluss an einen Vortrag, den ich im Frühling dieses Jahres in Lund in der Mendel-Gesellschaft hielt, mit, dass die *canina*-Rosen-Bastarde, die er in Kultur habe, und denen er früher auf Grund von Beobachtungen an den jungen Pflanzen völlig maternellen Charakter zugeschrieben habe, in mehreren Eigenschaften Spuren des Pollenspenders aufweisen. Die scheinbare maternelle Übereinstimmung muss demnach auch dort durch Dominanz bedingt sein. Gewisse Verschiedenheiten machen sich im Nachsommer bei meinen *R. rubrifolia*-Bastarden bemerkbar. Obwohl sie unter völlig gleichen Bedingungen aufgezogen worden sind, sind sie doch verschieden stark von Mehltau befallen. Zwei Pflanzen sind ganz intakt, drei sind sehr stark befallen, während die übrigen intermediären Befall zeigen.

Durch die obigen Resultate fallen alle Gründe fort, an das Vorliegen einer agamospermischen Samenbildungsweise bei den *canina*-Rosen zu glauben. Zwar ist es nicht bewiesen, dass Agamospermie nicht vorliegt. Man könnte sich ja das Vorkommen von Mixis nebst Agamospermie vorstellen, und dass diese nur durch 7 Chromosomen enthaltende Pollen aktiviert werden könnte. Die Gründe für eine solche Ansicht sind indessen nicht stärker als die Gründe dafür, dass jede beliebige normale Gattung als apomiktisch zu betrachten wäre. Alle die Eigentümlichkeiten bei den *canina*-Rosen können ja mit Hilfe der eigentümlichen Meiosis und der nachweislich vorliegenden balanzierten Heterogamie erklärt werden. Die *canina*-Rosen dürfen mithin nicht länger zu den agamospermisch sich vermehrenden Pflanzen gezählt werden.

Die nachgewiesenen Verhältnisse machen es notwendig, den systematischen Wert der innerhalb der Gruppe beschriebenen zahlreichen »Arten» anders als bisher zu beurteilen. Wenn die Konstanz nicht absolut und zweifellos in vielen Fällen nur eine scheinbare ist, verlieren diese »Arten» an Wert als systematische Einheiten. Der schwedische *canina*-Forscher MATSSON bemerkt des öfteren bezüglich der Konstanz der »Arten», dass sie in den wesentlichen Eigenschaften konstant seien. Die als wesentlich betrachteten Eigenschaften

sind wohl bedingt durch dominant wirkende Gene, die an die nicht geminibildenden Chromosomen gebunden sind. Durch diese Chromosomen werden die »wesentlichen Eigenschaften» auf den Nachkommen übertragen. Sämtliche Individuen, die durch nicht geminibildende Genome derselben Qualität zusammengehalten werden, als Repräsentanten einer distinkten Art zu betrachten, erscheint mir jedoch wenig zweckmässig. Die geminibildenden Genome können ja ganz verschieden sein. Auch wenn diese der äusseren Gestaltung des Individuums nicht in höherem Grade ihr Gepräge aufdrücken, können sie verschiedene physiologische Eigenschaften und Abspaltungen bedingen. Die Vorstellung von der Konstanz der *canina*-Arten ist indessen sicherlich übertrieben. Davon zeugt die eigentümliche Einteilung, die man bei den »Arten» vornehmen kann (vgl. ALMQUISTs Schema in der ersten Auflage von LINDMANS Flora). Sie können eingeteilt werden teils in Gruppen (*Afzelianae*, *Caninae*, *Villosae*, *Tomentosae* usw.), teils in gegen 30 Arttypen. Die meisten der Gruppen sind durch die meisten Arttypen repräsentiert. Die verschiedenen »Arten» können daher mit Hilfe eines Felderschemas bestimmt werden, in welchem die Gruppen waagerecht und die Arttypen senkrecht angeordnet sind. Mehrere der »Felder» enthalten vier Arten, eine *glauca*-Art (mit glatten, blaugrünen Blättern), eine *virens*-Art (mit glatten, rein grünen Blättern), eine *virentiformis*-Art (mit behaarten, rein grünen Blättern), eine *glauciformis*-Art (mit behaarten, rein blaugrünen Blättern). Innerhalb der meisten der so erhaltenen »Arten» würden dann kleinere Paralleleinheiten vorhanden sein, ALMQUISTs *angusti-*, *lati-*, *sub-*, *per-*, *hirti-*, *ob-*, *prae-* usw. -Formen. Die genannten Verhältnisse müssen wohl als wenigstens in hohem Grade genrekombinationsbedingt betrachtet werden. Eine zielbewusst betriebene Kreuzungsarbeit würde dies sicherlich aufzeigen.

Um den Bedingungen für die eigentümlichen meiotischen Verhältnisse bei den *canina*-Rosen näher beizukommen, habe ich im vergangenen Sommer *canina*-Rosen, vor allem *R. rubrifolia*, mit etwa 30 verschiedenen *Rosa*-Arten verschiedenen Polyploidiegrades gekreuzt. Bei den meisten Kombinationen habe ich ein positives Resultat in Form von, dem Aussehen nach, vitalem Samen erhalten. Die Samenqualität zeigt jedoch denselben Zusammenhang mit der Chromosomenzahl des Pollenspenders, wie er in meiner Zusammenstellung nachgewiesen wurde. Parallelversuche wurden mit *R. rugosa* ($2n = 14$), *gallica* ($2n = 28$) und *Sweginzowii* ($2n = 42$) als Mutterpflanzen angestellt. Im letzten Falle wurde ein völlig negatives Resultat erhalten, in den beiden anderen ein gutes. Um die Keim- und Befruchtungsfähigkeit des *canina*-Pollens zu untersuchen, wurde *rubrifolia*-, *rubiginosa*-, *glutinosa*-, *Seraphinii*- und *Afzeliana*-Pollen auf den Narben von *rubrifolia*, *rugosa*, *gallica* und *Sweginzowii* aufgetragen. Ein positives Resultat in Form von Samen wurde nicht mit *Sweginzowii*, aber mit den anderen Mutterarten in mehreren der Kombinationen erhalten.

Ystad, im Oktober 1941.

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ÁSKELL LÖVE: Polyploidy in *Polygonum Convolvulus* L. (s. lat.).

While studying the cytology of some species of the genus *Rumex* I made some counts of the chromosome numbers of some other members of the Polygonaceae. The purpose was partly to increase the knowledge of the somatic numbers of the Scandinavian species of the family, partly to find new types of variation in chromosome number within the polymorphous species complexes. In the family Polygonaceae only a few cases of polyploidy within the species are known. *Rumex scutatus* in England is tetraploid with $2n = 40$ chromosomes (FIKRY, 1930), but in Japan (NODA, 1926), Germany (JARETSKY, 1928) and in Sweden (LÖVE, unpubl.) it is to be found in the diploid state. In *Oxyria elatior* EDMAN (1929) reports a case of hexaploidy, and in the very polymorphous *Polygonum aviculare* L. (s. lat.) ANDERSSON (unpubl.) found varying chromosome numbers.

In the genus *Polygonum* only a few investigations of the chromosome numbers have been made. The most systematic studies of the cytology of a great number of species of this genus are those of JARETSKY (1928) and SUGIURA (1936). They found that we have the basic number $x = 11$ in some of the sections of the genus, and $x = 10$ in the others. In the section Tiniaria, which includes, among others, *Polygonum Convolvulus* L. (s. lat.), JARETSKY found the basic number $x = 10$, and the four species studied by him were all diploid. He found the diploid number in both the meiotic and the mitotic phases of plants of *Polygonum Convolvulus* L. from Kiel.

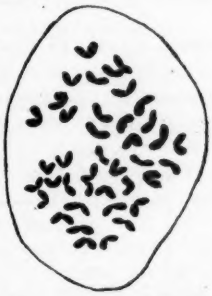

Fig. 1. Somatic chromosomes of *Polygonum Convolvulus* L. from Sweden. — × 3000.

Studying the chromosome number of Swedish material of *Polygonum Convolvulus* L. the present writer found the tetraploid number $2n = 40$ in the root-tip-cells (Fig. 1). The chromosomes are relatively small with a median or submedian centromere. No distinct chromosome morphological differences were observed between the members of the set.

My material originated from seeds collected in the neighbourhood of Lund. The plants were annual and flowered about two months after the germination of the seeds. The root-tips were fixed in LEWITSKY's 8 : 2 (cf. LÖVE, 1941) and all the slides were stained with gentian-violet.

As regards the systematics of the tetraploid form, it probably corresponds to *Polygonum Convolvulus* var. *sublaeve* ZAPALOW. (cf. ASCHERSON and GRAEBNER, 1908—1913). However, detailed studies of its taxonomic value have not yet been undertaken.

Studies of the chromosome numbers of plants with a wide geographical

distribution have often shown differences in the degree of polyploidy within different parts of the area. In most of the cases known the polyploid forms are to be found in alpine or boreal-arctic regions, while the diploid forms have a more southern distribution (cf. MÜNTZING, 1936). The tetraploidy in the Swedish form of *Polygonum Convolvulus* L. represents another case of this kind.

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HELGE JOHNSSON: Die Chromosomenzahl von *Carpinus betulus* L.

Mehrere Verfasser haben Chromosomenzahlen verschiedener Arten der Gattung *Carpinus* veröffentlicht. Dadurch sind die Chromosomenzahlen der meisten *Carpinus*-Arten bekannt worden. Die übereinstimmenden Ergebnisse dieser Untersuchungen sind, dass die Grundzahl der Gattung 8 ist und dass die untersuchten Arten Diploiden mit $n = 8$ sind. Unter den untersuchten Arten ist auch *Carpinus betulus*. Für diese Art hat WETZEL (1929) $n = 8$ konstatiert, welche Zahl von JARETZKY (1930) und WOODWORTH (1930 und 1931) bestätigt worden ist. Der letztgenannte Verfasser hat weiter gefunden, dass die Varietät *fastigiata* von *C. betulus* im Gegensatz zu der Hauptart $n = 32$ besitzt.

Eine im Institut für Forstpflanzenzüchtung in Schweden gemachte Untersuchung hat indessen das etwas überraschende Ergebnis geliefert, dass die Zahl, die WOODWORTH für *C. betulus* var. *fastigiata* angegeben hat, $n = 32$, allem Anscheine nach auch für die Hauptart gilt, was im Gegensatz zu den Angaben WETZELS, JARETZKYS und WOODWORTHS steht.

P.M.Z. von *C. betulus* und anderen *Carpinus*-Arten lassen sich ausserordentlich gut mit verdünntem Chromsäureformalin fixieren. Die Fixierungen wurden Ende Februar an abgeschnittenen Zweigen, nachdem diese einige Tage im Gewächshaus gestanden haben, leicht ausgeführt. Färbung mit Gentianaviolett gab gute Bilder. Mit Hilfe dieser Methode ist die Meiosis von 17 Bäumen von 7 verschiedenen schwedischen Standorten untersucht worden. Die Vorkommnisse, von denen 6 in Schonen und 1 in Uppland gelegen sind, sind teils spontan, teils gepflanzt. Fehler in bezug auf die systematische Bestimmung des Materials sind ganz ausgeschlossen, da *C. betulus* die einzige in Schweden spontan vorkommende *Carpinus*-Art ist.

Aus der Abhandlung WETZELs scheint hervorzugehen, dass er seine Bestimmung auf Material von einem Baum im Kieler Botanischen Garten gegründet hat. Dank gütigem Beistand von Herrn Professor G. TISCHLER habe ich Material von den zwei *C. betulus*-Bäumen, die dort wachsen, bekommen. Leider enthielt die Fixierung des eines Baumes keine P.M.Z.-Teilungen. Die Chromosomenzahl des anderen stimmte mit jener der schwedischen Bäume überein und war $n = 32$.

Die somatische Chromosomenzahl, $n = 64$, ist durch Bestimmungen in Wurzelspitzen von zwei Bäumen bestätigt worden. Material für diese Bestimmungen wurde von krautigen Stecklingen, die mit B-Indolbutylsäure behandelt worden sind, gewonnen.

Die Möglichkeit, dass es von *C. betulus* sowohl 16- wie 64-chromosomige Rassen gibt, ist zwar nicht völlig ausgeschlossen. Grosse Teile von *C. betulus* — jedenfalls aus den nördlichen Teilen des Verbreitungsgebietes — müssen jedoch durch die Chromosomenzahl $n = 32$ bzw. $2n = 64$ charakterisiert werden.

Gleichzeitig mit diesen Bestimmungen von *C. betulus* wurden auch P.M.Z. von *C. carpinizza* und *C. tshonoskoii* untersucht. Für diese beiden Arten wurden die Chromosomenzahlen zu $n = 8$ festgestellt.

Mit Hinblick auf das Vorkommen von 16-chromosomigen Arten von *Carpinus*, müssen *C. betulus*, oder jedenfalls Rassen von *C. betulus*, als oktoploid aufgefasst werden. Trotz dieser Verhältnisse haben sämtliche untersuchten *C. betulus*-Individuen eine völlig regelmässige Reifeteilung mit 32 Gemini in der I.M. ohne jede Spur von Multivalenten gezeigt. Und Univalente oder andere Störungen sind weder in der ersten noch in der zweiten Teilung angetroffen worden. Mit anderen Worten, die Reifeteilung des oktoploiden *C. betulus* ist auch regelmässiger als dies bei anemophilen Diploiden öfters der Fall ist. Desgleichen fehlt jede Spur einer »secundary association». Im Zusammenhang hiermit ist zu bemerken, dass die Chromosomen von *C. betulus* allerdings verhältnismässig klein sind; ihre Länge in der I.M. ist jedoch 3—4 Mal grösser als ihre Breite.

Von Interesse mit Rücksicht auf die Oktoploidie von *C. betulus* ist, dass die nahestehende Art, *C. carpinizza* (*C. betulus* var. *carpinizza*) diploid mit $n = 8$ ist. Der wesentlichste Unterschied zwischen diesen beiden Arten besteht darin, dass *C. carpinizza* kleinere Blätter hat als *C. betulus*. Ferner sei erwähnt, dass *C. betulus* die hochstämmigste *Carpinus*-Art ist; sie erreicht eine Höhe von 20 m, während keine der diploiden Arten eine Höhe von mehr als 15 m erreicht.

C. betulus ist die einzige *Carpinus*-Art Europas, abgesehen von kleineren Vorkommen von *C. orientalis* ($n = 8$) im südöstlichen Europa, und ist von Persien bis nach Mittelschweden verbreitet. Die übrigen Arten der Gattung sind in Nordamerika und Ostasien einheimisch.

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GERT BONNIER: The χ^2 linkage test.

When comparing frequencies expected according to some hypothesis with observed frequencies, a χ^2 is usually computed. The well-known definition of χ^2 is algebraically identical with the sum of the squared observed frequencies divided by the expected frequencies, from which sum the total frequency is substracted. Thus, in F_2 of a dihybrid cross between unlinked genes with normal 3 : 1 segregations, where the observed frequencies of the phenotypes *AB*, *Ab*, *aB*, *ab* are n_1, n_2, n_3, n_4 and $n_1 + n_2 + n_3 + n_4 = n$, we have by definition

$$\chi^2 = 16\left(\frac{n_1^2}{9n} + \frac{n_2^2}{3n} + \frac{n_3^2}{3n} + \frac{n_4^2}{n}\right) - n \qquad (1)$$

To this value there are 3 corresponding degrees of freedom. Now consider the 3 biological processes at work, viz. two 3 : 1 segregations and absence of linkage. For the $A : a$ segregation the 2 observed frequencies are $n_1 + n_2$ and $n_3 + n_4$, and the expected ones are $\frac{3n}{4}$ and $\frac{n}{4}$. After some simplifications this gives as the part of χ^2 corresponding to this segregation:

$$\chi_A^2 = \frac{(n_1 + n_2 - 3n_3 - 3n_4)^2}{3n} \qquad (2)$$

and correspondingly for the $B : b$ segregation:

$$\chi_B^2 = \frac{(n_1 - 3n_2 + n_3 - 3n_4)^2}{3n} \qquad (3)$$

Each of these two parts of χ^2 has one degree of freedom. Taking for granted that the sum of the three parts of χ^2 must equal the total χ^2, we may find the part, χ_L^2, corresponding to the absence of linkage by substracting $\chi_A^2 + \chi_B^2$ from χ^2, from which

$$\chi_L^2 = \frac{(n_1 - 3n_2 - 3n_3 + 9n_4)^2}{9n} \qquad (4)$$

This value, which is usually adopted, does not, however, conform to the general definition of a χ^2. The two observed frequencies corresponding to the hypothesis of absence of linkage are $n_1 + n_4$ and $n_2 + n_3$, i. e. the parental combinations and the recombinations. If there are normal 3 : 1 segregations the two expected frequencies are obviously $\frac{5n}{8}$ and $\frac{3n}{8}$, from which the χ^2 of the linkage test turns out to be

$$\chi_k^2 = \frac{(3n_1 - 5n_2 - 5n_3 + 3n_4)^2}{15n} \tag{5}$$

If the $A : a$ and $B : b$ segregations do not conform to the 3 : 1 hypothesis, the expected proportion of parental combinations may be called p. Thus if $q = 1 - p$, the more general formula of the χ^2 for absence of linkage is

$$\chi_K^2 = \frac{(qn_1 - pn_2 - pn_3 + qn_4)^2}{pqn} \tag{6}$$

The value of p may be estimated in the following manner. Absence of linkage rests on the hypothesis that one half of the F_1 gametes are parental combinations, *AB* and *ab*, and one half recombinations, *Ab* and *aB*. Now let the proportion of *AB-gametes* be s and of *Ab-gametes* be t. Going through the F_2 checkerboard it is found

expected proportion in F_2 of phenotype $AB = 2s - s^2 + t - 2t^2$ (7)

» » » » » » $Ab = t^2 + t - 2st$ (8)

» » » » » » $aB = \frac{3}{4} - s - 2t + t^2 + 2st$ (9)

» » » » » » $ab = \frac{1}{4} - s + s^2$ (10)

By adding the expected proportions of phenotypes *AB* and *ab* we get

$$p = s + t - 2t^2 + \frac{1}{4} \tag{11}$$

Thus, if we can estimate s and t, p is also estimated. Now the observed proportion of the phenotype A is $\frac{n_1 + n_2}{n}$ and that of phenotype B $\frac{n_1 + n_3}{n}$. Following the principle of maximum likelihood, the expected proportions of these phenotypes are to be equalized to the observed proportions, from which

$$2(s + t) - (s + t)^2 = \frac{n_1 + n_2}{n} \tag{12}$$

$$(s - t) - (s - t)^2 + \frac{3}{4} = \frac{n_1 + n_3}{n} \tag{13}$$

Solving s and t, it is found that

$$s = \frac{3}{4} - \frac{1}{2}\left(\sqrt{\frac{n_2 + n_4}{n}} + \sqrt{\frac{n_3 + n_4}{n}}\right) \tag{14}$$

$$t = \frac{1}{4} + \frac{1}{2}\left(\sqrt{\frac{n_2 + n_4}{n}} - \sqrt{\frac{n_3 + n_4}{n}}\right) \tag{15}$$

By substituting in (11) the proportion p is estimated. If $A : a$ segregates normally in 3 : 1 but not $B : b$, $s + t = \frac{1}{2}$. If the converse is the case, $s = t$. This easily gives as estimation of p,

when $A : a$ segregates in 3 : 1 but not $B : b$

$$p = \frac{3}{4} - \frac{n_2 + n_4}{2n} \tag{16}$$

when $B : b$ segregates in 3 : 1 but not $A : a$

$$p = \frac{3}{4} - \frac{n_3 + n_4}{2n} \tag{17}$$

χ_K^2 vanishes for all frequencies which are proportional to the values (7), (8), (9), (10), i. e. when there is a numerically exact absence of linkage. Now supposing that there is such a complete absence of linkage, i. e. supposing that $\frac{n_1}{n}$, $\frac{n_2}{n}$, $\frac{n_3}{n}$, $\frac{n_4}{n}$ are equal to the values (7), (8), (9), (10), this may be substituted in (4), which in this case gives

$$\chi_L^2 = \frac{16n}{9}(s - t)^2 [2(s + t) - 1]^2$$

But this value vanishes only if either $s + t = \frac{1}{2}$ — when $A : a$ segregates in 3 : 1 — or $s = t$ — when $B : b$ segregates in 3 : 1. Thus, to ensure that the usually adopted χ^2-linkage value (4) is to equal zero, it is not enough that there is a complete absence of linkage, it is also necessary that one or both of the segregations must at the same time follow the 3 : 1 hypothesis. In other words, χ_L^2 is not a test of the single biological process, absence of linkage, but of at least two different processes, absence of linkage and a 3 : 1 segregation.

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A. AND G. MÜNTZING: Recent results in *Potentilla*.

As reported in previous publications (MÜNTZING, 1928, 1931), the *Potentilla* species *argentea, collina, Tabernæmontani* and *Crantzii* are pseudogamous and comprise biotypes with different multiples of the basic chromosome number seven. New chromosome counts were undertaken in 53 strains of *P. argentea* L. from 37 different localities in Sweden and Norway. 37 of

these strains were diploid ($2n = 14$), 12 hexaploid ($2n = 42$), 3 tetraploid ($2n = 28$) and 1 pentaploid ($2n = 35$). All strains so far observed were completely uniform, but always differed from each other in various morphological and physiological respects. This constancy must be due to apomixis, and has already been demonstrated by experimental work in 6 *argentea* types (MÜNTZING, 1928). Further evidence of the same kind was obtained from the pentaploid *argentea* biotype, which combines complete constancy with an odd multiple of seven and hence with an irregular meiosis.

A morphological analysis demonstrated that the diploids correspond to *P. argentea* MARKL., and the hexaploids to *P. impolita* WAHLENB. The tetraploid biotypes are morphologically intermediate between the diploids and hexaploids. The pentaploid biotype cannot morphologically be distinguished from the hexaploids. The diploid biotypes seem to have a more northern area of distribution than the hexaploids. The two groups also differ in periodicity, rust resistance, seed and pollen size and in average pollen fertility (cf. A. and G. MÜNTZING, 1941).

Meiosis in the pentaploid *argentea* biotype was studied. At first metaphase 11 different configurations were observed, the most frequent configuration being $2_{III} + 12_{II} + 5_{I}$. The most plausible explanation of the observed mode of chromosome pairing is that the pentaploid biotype in question contains several more or less homologous genomes. Four of these genomes generally pair as bivalents or form an occasional quadrivalent. The fifth genome may in rare cases occur as 7 univalents, but more frequently some of these chromosomes form trivalents with homologues in the other genomes. The occasional occurrence of 15 bivalents + trivalents may be due to intragenomatic pairing.

Two foreign deviating *argentea* types have been studied. The first of these is an octoploid ($2n = 56$), apomictic biotype, characterized by great vigour, lateness and strictly perennial duration of life. Morphologically it is a super-*impolita* type. The other deviating biotype, *A—C*, obtained from Switzerland, is a diploid and partially sexual strain. In contrast to the diploid Scandinavian *argentea* types *A—C* is strictly perennial and is also morphologically divergent in several respects. Thus, it is quite clear that the names *P. argentea* MARKL. and *P. impolita* WAHLENB. cover only part of the biotypes belonging to the collective species *P. argentea* L.

In crosses with hexaploid biotypes of *P. Tabernæmontani* and *collina* the *A—C* strain has only given maternal offspring. On the other hand, in crosses with other members of the *argentea* group true hybrids were almost exclusively produced, only a small proportion of maternal plants being formed. When crossed with a diploid Scandinavian *argentea* biotype (as the male parent) the *A—C* strain, as expected, gave diploid hybrids ($2n = 14$). This was controlled in 28 hybrid plants, no deviating chromosome numbers being observed. The hybrids between *A—C* and hexaploid *argentea* (= *impolita*) should have $2n = 28$. An interesting feature, however, is that almost all hybrids of this kind (involving 3 different hexaploid biotypes) were *pentaploid* ($2n = 35$). Of 30 plants produced from these crosses 28 were found to have $2n = 35$ or $2n = \pm 35$, one single plant having the expected chromosome number $2n = 28$ and one plant being diploid with $2n = 14$. Thus, in

crosses with other diploid *argentea* types the reduced ovules of the mother strain, *A—C*, function and give viable zygotes. In crosses with hexaploid biotypes, on the contrary, the reduced ovules are seldom fertilized or seldom give viable zygotes, the latter alternative being the more probable one. Viable seeds are almost exclusively obtained from the union of unreduced ovules and reduced male gametes ($14 + 21$). — This case is evidently quite analogous to the results obtained by NORDENSKIÖLD (1937) in crosses between diploid *Phleum nodosum* and hexaploid *Phleum pratense*. Other cases of a similar kind have been reviewed by MÜNTZING (1936, pp. 329—330).

The pentaploid hybrids in our material have not yet flowered. The diploid hybrids between *A—C* and diploid Scandinavian *argentea* biotypes were found to be rather sterile and to give a typical species cross segregation in F_2. Thus, *A—C* and the Scandinavian diploids must be regarded as specifically different. Some F_2 plants were triploid and had a high frequency of trivalents at meiosis.

Another, almost completely sexual strain of diploid *argentea* was obtained, probably as the result of a spontaneous cross between the *argentea* biotype *A—G* and the partially sexual strain, *A—C*, discussed above. This new sexual strain was utilized for the production of various hybrids. One spontaneous pentaploid hybrid (probably with hexaploid *argentea* as the male parent) was found to be purely sexual, and gave a highly variable progeny. In crosses with *P. Tabernæmontani* only a few maternal plants were obtained. Crosses with hexaploid *argentea* resulted in 8 maternal plants, 2 hybrids with $2n = \pm 35$ and 1 plant having $2n = \pm 43$. Thus, also in this case viable hybrid zygotes were only obtained from unreduced ovules or, in one case, from an ovule with approximately 22 chromosomes. These results, and especially the evidence obtained from the crosses with *A—C*, demonstrate that the result of a cross in our *Potentilla* material is not only determined by the degree of sexuality of the mother strain but also by the chromosome number and the genetical constitution of the male parent.

Hybrids between *P. argentea* and *opaca* were produced by using the sexual *argentea* strain derived from *A—G* as the female parent. Most F_1 hybrids were diploid, like the parents, and were found to have an almost entirely regular meiosis but poor fertility. Some of the F_1 hybrids were triploid, having two *argentea* genomes and one *opaca* genome. The majority of the hybrids were extremely vigorous, but a certain *argentea* plant, though itself normally developed, gave hybrids with a very poor vigour.

A few true hybrids between the otherwise apomictic *P. Tabernæmontani* biotypes *T—B* ($2n = 84$) and *T—A* ($2n = 42$) were obtained, *T—B* being the female parent in the successful cross. The hybrids ($2n = 63$) gave only apomictic progeny, the chromosome number invariably being 63 in 73 F_2 plants examined. These plants were in all morphological respects identical with the F_1 plants and did not show any segregation at all. Thus, the cross resulted in the sudden creation of a new constant biotype with a new multiple of the basic chromosome number of the genus.

Another similar case is represented by the spontaneous production of a *P. collina* plant with doubled chromosome number. The biotype in question, *C—A*, is hexaploid ($2n = 42$) and was previously found to be quite constant

and strictly pseudogamous (MÜNTZING, 1928). In an otherwise uniform plot of *C—A* a single individual was observed to have typical gigas characters and an increased chromosome number. The exact chromosome number of this plant has not yet been determined, but 6 daughter plants obtained after isolation have $2n = 84$. Under such circumstances the mother plant must certainly also have $2n = 84$, thus being autotetraploid in relation to the normal plants of the strain. Two further daughter plants, however, were found to have the chromosome numbers 42 and 43 respectively. This indicates that a small proportion of »haploids» are produced in this new strain just as in similar cases in the genus *Poa* (cf. MÜNTZING, 1940).

The genotypical basis of apomixis and the causes of polymorphism in *Potentilla* are discussed in a recent paper (A. and G. MÜNTZING, 1941). In the same paper much of the evidence briefly summarized here is considered in detail. The present report also comprises some new results obtained in the summer of 1941.

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ÅKE GUSTAFSSON: Cross-experiments in the genus *Rosa*.

In 1930 the present writer began his studies on the modes of propagation in the genus *Rosa*, most closely in connexion with rather extensive investigations of species-problems in *Rubus*. In two short papers in 1931 it was proved by means of emasculation experiments that no autonomous apomixis occurs in the *canina*-roses. Artificial crossings were performed, and during the following years a great number of offspring plants arose from such pollination experiments. Here a brief survey of the crosses and some immediate genetical data will be presented. A full description of the genetics and cytology of F_1- and F_2-individuals will be published later.

The following cross-combinations were tried: *R. canina* L. var. *blandaeana* CREP. × *rugosa* THUNB., *R. canina* L. var. I × *rugosa* THUNB., *R. canina* L. var. II × *rugosa* THUNB., *R. rubiginosa* L. × *rugosa* THUNB., *R. elliptica* KELL. × *coriifolia* FR., *R. glauca* POURR. × *pomifera* HERRM., *R. mollis* CHRIST. × *pomifera* HERRM., *R. tomentella* LEM. × *rubrifolia* VILL., *R. rubiginosa* L. × *canina* L. var. II, *R. canina* L. var. II × *rubiginosa* L., *R. rubri-*

folia VILL. × *rubiginosa* L., and *R. rubiginosa* L. × *rubrifolia* VILL. In addition, a great many pollinations with *R. rugosa* THUNB. as mother have been carried out, but so far they have failed.

Offspring plants were obtained in the combinations *R. canina* var. *blandaeana* × *rugosa* (1932: 1 and 2), *R. canina* var. I × *rugosa* (1934: 1), *R. canina* var. II × *rugosa* (1934: 4), *R. rubiginosa* × *rugosa* (1932: 5 and 1934: 2 and 3), *R. canina* var. II × *rubiginosa* (1934: 5), *R. rubiginosa* × *canina* var. II (1934: 6), *R. rubrifolia* × *rubiginosa* (1935: 13), and *R. tomentella* × *rubrifolia* (1932: 4). In the different crosses there are still alive about 35, 5, 2, 19, 13, 6, about 50, 1 and 1 plants respectively. No final account of the results can be given here, as some combinations require F_2-generations in order to ascertain the mode of propagation.

1932: 1 and 2 as well as 4 and 5 flowered for the first time in 1937—1938, 1934: 1, 2, 3, 4, 5 and 6 in 1939—1940—1941 and 1935: 13 in 1941. The oldest plants are by now seven years of age. F_2-plants, arisen in isolation experiments, were raised from 1934: 6 exclusively, but here a great many two-years seedlings already exist.

In the cross-combination 1934: 1 (*R. canina* var. I × *rugosa*) five plants arose, all of them typical intermediate forms between the parents. The most striking feature is the intensely red petals, redder than in the mother, but not so deep-red as in the father. The spines are numerous, unequal and partly exceedingly strong, the leaves thick and leather-like, but in shape and serrature they agree with those of the mother. The hips are extremely large, in one individual larger than in both parents, with a content of ascorbic acid well above that of the mother and certainly on a par with that of the father. The sterility is rather pronounced, despite the well-developed hips, 6,0, 8,9, 4,4, 7,0 and 5,8 nuts per hip being formed as against 28,9 in *R. canina* var. I and 27,2 in *R. rugosa*.

All five specimens have a different morphological appearance. Plant 2 is especially strong, plant 4 rather weak, the leaves of this plant moreover being yellowish-green. Plant 3 differs by having smaller and narrower end-leaflets. Further, the hip-size varies considerably. The breadth is 19,2, 18,6, 14,8 14,0 and 17,9 mm. respectively, the length being 27,4, 28,7, 27,4, 24,0, 25,9 mm. Consequently, plant 3 has remarkably long and narrow hips as compared with plants 1 and 2, and the hips of plant 1 are even more rounded than those of plant 2.

We may conclude that the mother-plant of this cross was sexual.

In the cross *R. canina* var. *blandaeana* × *rugosa* a great many progeny plants were obtained. In the beginning they were recorded as maternal (cf. the conclusion in GUSTAFSSON, 1937), but now that they have flowered and fructified several times and attained full-grown size, there cannot be any doubt as to their hybrid condition. Certainly the leaf-shape and serrature are much the same as those of the mother, but the spines are strong, rather numerous and unequal, the petals pinkish-red, the sepals somewhat fleshy and more or less persisting, all characters distinguishing them from the mother. The hips are larger than in the parents, the breadth and length being on an average 14,8 and 24,8 mm. (7 specimens, 77 hips), against 14,2 and 23,8 in *R. canina* var. *blandaeana* (10 hips) and 21,4 and 18,4 in *R. rugosa* (5 hips).

Very conspicuous is the sterility. Seven specimens showed, on an average, 4,4 nuts per hip as against 21,5 and 41,0 in the parent species. Like in the preceding case, the content of ascorbic acid is twice or three times that of the mother, but somewhat lower than in the father.

Some small variation in regard to spines, leaf- and hip-size seems to exist, but no definite conclusion can be arrived at until the second generation is raised.

Even this cross-combination proves that the mother-type — at least when pollinated with *R. rugosa* — is purely sexual. The properties of the mother prevail, however, to a greater extent than was the case in the former instance.

Of the hybrid *R. rubiginosa* × *rugosa* a great many individuals were obtained. In the series 1934: 2 and 3 no maternal specimens arose, but the leaf-shape, the serrature and the glands are reminiscent of the mother. The extremely dense, unequal spines and prickles prove the successful union of the two types of gametes. The shrubs are strong and rapid-growing, the hip-formation good but the sterility is nevertheless rather pronounced. Five specimens of 1934: 2 produced 5,6, 5,3, 2,6, 8,8 and 5,6 nuts per hip, as against 19,3 and 27,2 in the parents. A distinct variation was observed, although most of the specimens were alike.

In the series 1932: 5 the sterility was even higher, as most flowers fall off after opening. This was especially noticed in the years 1938 and 1939, when almost no hips developed. In 1941 the hips arose more frequently, although not as frequently as in the crosses 1934: 2 and 3. The number of seeds is lower than in the other series, viz. 5,1, 4,0, 3,3 and 2,9 nuts per hip in four specimens as compared with 17,1 and 41,0 in the parents. The entire progeny, except for one plant, was rather uniform and in many morphological characters resembled the crosses 1934: 2 and 3. The exceptional plant is probably maternal, arisen either by pseudogamy or maybe by incomplete emasculation (19,4 nuts per hip).

Thus *R. rubiginosa* (two different shrubs) is also sexual in combination with *R. rugosa*. One out of about 30 individuals was maternal, probably due to some experimental error.

A further *rugosa*-cross remains (1934: 4). Here the situation is more complex, since one specimen shows no signs of the father, but yet without being maternal, and since the other specimen is probably a true hybrid with a great prevalence of the mother's characters. The former was entirely sterile in 1941, no hips being formed, although it flowered abundantly. The latter plant agrees in respect to sterility, hip-properties and content of ascorbic acid with the *canina-rugosa* hybrids mentioned previously. Cytological studies as well as progeny controls will at once elucidate the state of things.

The series 1934: 5 (*R. canina* var. II × *rubiginosa*) consists of six individuals, evidently being true F_1-hybrids but differing more or less from one another. The hips are rather large and well-developed, in two instances larger than in both parents, although the number of nuts is appreciably decreased (2,6, 3,1, 4,3 and 4,9 in four individuals as against 16,2 and 19,3 in the parents). Plant 6 deviates conspicuously from the others, and in several properties it approaches the father. Obviously *R. canina* var. II is sexual in

combination with *R. rubiginosa*, and gametes of these two species together form a heterogenous F_1.

There still remain the combinations of *R. tomentella* × *rubrifolia*, *R. rubiginosa* × *canina* var. II and *R. rubrifolia* × *rubiginosa*. In the first instance no trace of the singular pollen-species can be noticed. The cause may be a high degree of prevalence, but it may also be due to the occurrence of pseudogamy or incomplete emasculation. In the second case the mother-properties predominate and the fertility is normal (except in one specimen). Nevertheless it appears as if this series, too, consists of true hybrids. A daughter generation, now in its second year, will in a few years clear up the matter. The single plant of *R. rubrifolia* × *rubiginosa* is too young to allow definite conclusions.

These data indicate no doubt that the *canina*-roses are entirely sexual, at least in special combinations. In fact, no absolutely sure case of pseudogamy was recorded, the few maternal plants occurring probably being due to the prevalence of the mother genes or incomplete emasculation. Slowly F_2-data will accumulate and will fully solve the intricate problem.

Pseudogamy being absent or very rare, the *canina*-roses will form an example of a great and complex natural group, like the *Oenotheras*, having acquired a special cytological mechanism. They still possess the quality of constancy, thereby preserving the valuable properties, but also the ability of variation by hybridisation and segregation. The constancy is maintained by a special system of heterogamy (BLACKBURN and HARRISON, 1921; GUSTAFSSON, 1931; DARLINGTON, 1932 and FAGERLIND, 1940). Finally, in the pseudogamous tetraploid mass of European blackberries a third case of a special equilibrium system occurs, here conditioned by the sudden origin of sexuality in hybrids and the gradual recovery of pseudogamy in segregation.

In fact, the above conclusion was drawn by GENTCHEFF and GUSTAFSSON in 1940 (p. 110).

Svalöf, Sweden, November 1941.

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G. ÖSTERGREN: Note on the acetocarmine method.

During a study on the cytology of *Anthoxanthum* the present writer has also made some experiments with the acetocarmine method. The technique of MCCLINTOCK to fix the anthers in the CARNOY fixative alcohol—glacial acetic acid 3 : 1 before smearing in acetocarmine gives a considerably better fixation than smearing directly. To make it possible to dinstinguish at once after smearing between preparations with divisions and without, I had the idea of *staining the anthers as a whole over night or for a few days in iron aceto-carmine*, and after that to smear. The idea worked quite well in practice. I think this treatment at *room temperature* to be less deleterious than treating with boiling acetocarmine, as recommended by some workers. When boiling, the liquid is also often adulterated by precipitates of carmine or iron-carmine; this is, of course, avoided by staining at room temperature. The fixing in CARNOY should last for 12—24 hours; a prolonged treatment has definitely deleterious effects, the cells shrinking and the spindle being exaggerated. Transference to alcohol between fixing and staining also decreases the quality of the result. It should be noted that an anther which has been treated with alcohol-acetic acid and acetocarmine, as described here, has *quite other properties when submitted to smearing* than a fresh anther. A unicellular layer of a large number of p.m.c:s and tapetum cells is much more easily obtained after such a treatment. A quite satisfactory adherence of the cells to the slide when making the preparation permanent was obtained by using slides prepared with albumen, which had been dried over a flame. Albumen-prepared slides have been used in smearing by BAUER and by BRIDGES (1937) and EIGSTI (1937). The smearing was performed by putting a cover glass on the anther (in a drop of acetocarmine on the slide) and pressing on the cover with a match. The cover glass should not be allowed to slip horizontally, and is to this end kept steady during smearing by pressing on it on two opposite sides with blotting-paper, which also absorbs the excess of acetocarmine. To protect the fingers from acetocarmine a piece of rubber sheet is put between the paper and the finger. The preparations were made permanent chiefly in accordance with the method of BUCK (1935).

The preparations obtained in *Anthoxanthum odoratum* by the method described are superior to those obtained in the usual way after fixing in chrome-acetic-formalin and staining with gentian violet (preparations sectioned with a microtome). The contrast in stain between chromosomes and plasm is not so strong as in gentian violet preparations but the chromosome configurations are nevertheless more easily analyzable. If the chromosomes are swollen from the influence of the acetic acid, it is not to such an extent as to influence the analyzability, at least not in an unfavourable way. I have the impression that they are not so swollen as they usually are in acetocarmine preparations produced directly from fresh material (not previously fixed in CARNOY).

I have also tried to make permanent preparations with another method. The anthers, stained as above, were dehydrated through increasing concentrations of alcohol and transferred to alcohol + xylol and to xylol. They were then put in Canada balsam and smeared. A preparation obtained in this

way, of course, should be permanent at once. The effort was unsuccessful, because the anthers had been hardened through dehydration and transference to xylol, and were not at all suitable to smear.

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G. VON ROSEN: Eine physiologische Wirkung der Blütengrösse bei *Pisum*.

In den Artkreuzungen zwischen verschiedenen theoretischen Linien von *Pisum sativum* L. und *Pisum abyssinicum* BRAUN, die Verf. ausser anderen Artkreuzungen in derselben Gattung ausgeführt hat (VON ROSEN, 1942), ist eine eigentümliche Erscheinung beobachtet worden.

Die Breite der Fahne, als ein Mass der Grösse der Blüte, schwankt bei ungefähr 30 benutzten *Pisum sativum*-Linien etwa 2,0—2,7 cm. Die Fahne von *Pisum abyssinicum* ist indessen nur 0,8—1,5 cm breit. Mit der Blütengrösse ist aber eine physiologische Eigentümlichkeit gekoppelt. Die individuelle Blüte des früheren Typus bleibt, abhängig von der Witterung, drei bis vier Tage geöffnet, schliesst aber während dieser Zeit jeden Abend bei der Dämmerung die Perigonblätter und öffnet sie wieder jeden Morgen. Die *abyssinicum*-Blüte dagegen blüht nur ein bis zwei Tage, schliesst sich auch am Abend, nur noch früher, und tut dies überdies sobald der Himmel sich stark bewölkt oder bei Regenwetter. Die kleine *abyssinicum*-Blüte benimmt sich in dieser Hinsicht ganz anders als die *sativum*-Blüte.

In F_1 und ebenso in F_2 der Kreuzungen zwischen diesen beiden Arten ist die Blütengrösse ungefähr intermediär (im Durchschnitt 2,0 cm), obschon die Variationsbreite innerhalb der einzelnen Familien viel grösser ist als im Vergleich mit den Elternlinien. In F_1 benehmen sich alle und in F_2 beinahe alle ganz wie die *sativum*-Eltern. Jede Blüte lebt also 3 bis 4 Tage und schliesst die Perigonblätter nur zur Dämmerung. Eine einzige Kreuzung wich von den übrigen ab. (Sie hat sich auch in verschiedenen genotypischen Hinsichten als eigentümlich erwiesen, was später veröffentlicht werden soll.) Die Blüten zeigten nämlich in diesem Fall Fahnen von im Durchschnitt 3,2 cm Grösse, die so ausgespannt waren, dass sie oben in der Mitte oft einrissen. Die Variation der Grösse war erheblich: 2,0—3,5 cm Breite. Dazu kam, dass dieselbe Blüte 3—5 Tage blühte, ohne sich vor dem Absterben ein einziges Mal zu schliessen, weder nachts noch bei Regen.

Aus der Untersuchung dieser Erscheinung hat sich ergeben, dass die Blütengrösse genetisch bedingt ist, aber auch dass sie von der Vitalität der Pflanze sehr abhängig ist, was ja schon lange bekannt war.

Diese Beobachtungen scheinen also zu zeigen, dass Blüten mit einer

Fahne von ca. 1 cm Breite kurz blühen und dazu gegen Lichtveränderungen sehr empfindlich sind. Die von 2 cm Grösse blühen länger und schliessen die Perigonblätter nur abends, während die von 3 cm Grösse ebenso lange wie die letztgenannten, oder möglicherweise noch etwas länger blühen, sich aber zwischen Aufblühen und Verwelken niemals schliessen. *Demgemäss scheint zwischen der genetisch bedingten Blütengrösse und der physiologischen Eigenschaft »Öffnen und Schliessen» der Perigonblätter mit dem Lichte bei Pisum ein Zusammenhang zu bestehen.*

Hilleshög, im Oktober 1941.

DORIS LÖVE: A tetraploid, dioecious *Melandrium* found in nature.

In recent years the studies of sex determination in polyploid dioecious species have shown that in the vegetable kingdom there are at least two types of sex determination, both based on the occurrence of sex chromosomes. In *Rumex Acetosa*, ONO (1935) found that the triploids present in nature were intersexes or females because the balance between the female-determining X-chromosome and the male-determining autosomes has been disturbed. In induced tetraploids of *Melandrium album*, however, WESTERGAARD (1940) showed that polyploids of this species were dioecious because the sex is determined by a strong male element in the Y-chromosome. WESTERGAARD also mentions that, theoretically, a tetraploid »race» of this species should be able to exist in nature and have a normal sex ratio.

For some years the present writer has been carrying on cytological work in the genus *Melandrium*, especially concerning the *M. rubrum* complex in Scandinavia. These studies have shown that in the main type of the species we have the diploid number $2n = 24$, as found by BLACKBURN and others (cf. LÖVE, 1940), but in one spontaneous plant the tetraploid number $2n = 48$ was found.

Studies of the taxonomy of the tetraploid plant show that it belongs to *Melandrium crassifolium*, described by FRIES in 1857. Investigations of the pollen diameter, etc. of herbarium material of this and other Scandinavian types of the collective species *M. rubrum* indicate that *M. crassifolium* and *M. rubrum* ssp. *lapponicum* (SIMMONS, 1907) are identical. These tetraploid forms seem to be those described by TURESSON (1925) as a coast ecotype and partly also as a subalpine ecotype of *M. rubrum*.

Institute of Genetics, University of Lund, Nov. 1941.

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G. ÖSTERGREN: Chromosome numbers in *Anthoxanthum*.

The chromosome numbers previously known in *Anthoxanthum* are: *A. odoratum*, $2n = 20$, KATTERMANN (1931), AVDULOW (1931), BÖCHER (1938), PARTHASARATHY (1939); HUNTER (1934) found $2n = 20 + 6ff$, I think the nature of his fragments is rather doubtful, however. *A. aristatum*, $2n = 10$, AVDULOW (1931).

Meiosis has been studied in *A. odoratum* only (KATTERMANN, l. c. and PARTHASARATHY, l. c.).

I have made my chromosome number determinations on root tips fixed in chrome-acetic-formalin, sectioned with a microtome and stained with gentian violet.

Annual species.

A. aristatum BOISS. Wild material from Coimbra, Portugal; typical in appearance. $2n = 10$ and $10 + 2ff$, $10 + 3ff$. Meiosis: 5_{II}. Material from three botanical gardens; not typical, shows characteristics similar to *A. ovatum*. $2n = 10$. Meiosis: 5_{II}.

A. ovatum LAG. Wild material from Kenitra, Mamora forest, Morocco. Adult plants not seen by me as yet. $2n = 10$. Meiosis not studied.

Perennial species.

A. odoratum L. Wild material from alpine and subalpine localities in Norway, Northern Sweden and Switzerland. $2n = 10$. Meiosis not studied.

A. odoratum L. Wild material from Scandinavian lowland and Iceland (not alpine conditions), material from botanical gardens. $2n = 20$. A few plants had $2n = 21$; this variation from plant to plant is probably due to the meiotic irregularities caused by autopolyploidy, as in *Dactylis glomerata* (MÜNTZING, 1937). (I have the notion that the two chromosomal races in *A. odoratum* may form a case rather analogous to the two species of *Dactylis*, *D. glomerata* and *D. Aschersoniana*.) Meiosis: A high frequency of quadrivalents and a few configurations of even higher order. Inversion bridges. My observations agree on the whole with those of KATTERMANN and PARTHASARATHY. The configurations are explicable as due to autopolyploidy accompanied by structural hybridity (segmental interchange, inversion heterozygosity).

A. amarum BROT. Wild material from Coimbra, Portugal; typical in appearance. $2n = 80$ (16-ploidy). The determination of such high chromosome numbers must evidently be somewhat uncertain; still it was possible to make quite certain the existence of a variation in chromosome number from

plant to plant. Probably this variation is due to autopolyploidy as supposed for *A. odoratum*. I hope to be able to study meiosis of this species soon.

The two chromosome numbers in *A. odoratum* are connected with a different ecological and geographical distribution, and also to some extent with morphological differences. The diploid is usually not so tall as the tetraploid, it has narrower leaves and smaller floral parts (spikelets). The awns usually protrude more than in the tetraploid. The colour of the ripe panicles is often warmer (more like gold) in the diploid, those of the tetraploid being paler yellow. Still it is not possible by mere inspection to distinguish the two »races», as there is a great variation in both of them. As the hybrid between them is a triploid, there will evidently be an efficient sterility-barrier between them. As far as I can see they have all properties necessary to make them different species, except, perhaps, that of sufficient morphological differences.

In this connection the way we look at the species concept is rather important, viz. if we think species exist only in the mind of man, or if we think they have an existence in nature quite independent of man. If we take this last position, we cannot, indeed, make our capacity or incapacity of morphologically recognizing them the crucial point in species delimitation, especially as this capacity may vary highly from one person to another. Anyhow, this is a delicate problem, and I think I will not decide on the systematic position of these »chromosomal races» in *Anthoxanthum* as yet. Nor have I taken up any position as regards the taxonomic interrelations of the other species of the genus, e. g. if *A. aristatum* BOISS. and *A. Puelii* LEC. et LAM. really are different species.

I have also had some success with artificial species hybridisation in *Anthoxanthum*. The meiosis in the triploid hybrid *A. aristatum* × *odoratum* has been observed by me.

A more extensive paper on this subject will be published later on.

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ALBERT LEVAN: The macroscopic colchicine effect — a hormonic action?

If a growing plant organ, as, for instance, a root tip, is treated with colchicine, it responds with the formation of a very characteristic tumour (the c-tumour reaction; LEVAN, 1938). A similar reaction is also obtained by certain growth substances in suitable concentrations. The present writer has shown (1939) that the cytological mechanism connected with the latter phenomenon is entirely different from the colchicine effects. While the growth substances primarily bring about a swelling of the cortical cells, which in its turn causes an endomitotic duplication of the chromosomes, colchicine acts on the spindle, producing restitution nuclei. It has therefore been generally assumed that the c-tumour reaction is a secondary phenomenon caused by the increase in chromosome number.

Recent investigations on colchicine effects in x-rayed plant organs show, however, that this view is erroneous. Dormant root primordia of *Allium fistulosum* bulbils were x-rayed with a heavy dosage. The root tips were then germinated in water and in different concentrations of colchicine solutions. While the roots growing in pure water were normal in appearance, all the colchicine treated roots formed very conspicuous c-tumours. The cytological investigation showed that the early growth of the roots took place entirely by cell enlargements and not by mitoses, the first real mitoses being found on the fifth day after the onset of germination. By that time very prominent c-tumours had already been formed. It was clearly seen that the cells of the control roots stretched mainly in a longitudinal direction, while the cells of the colchicine treated roots had their greatest increase transversally. The maximal swelling occurred in the cortical tissue immediately above the tip meristem. The growth of this tissue was occasionally so extreme that the central vascular tissue was entirely ruptured by the tension, resulting a great central cavity. As no mitoses were ever found in the cortex, it could not be decided whether the swelling of the cells involved endomitotic divisions of the nuclei.

It is concluded that colchicine brings about a similar increase in cell size as, for instance, naphthalene acetic acid, the enlargement of the cells occurring parallelly *and not secondarily* to the more conspicuous effect of colchicine on the mitotic mechanism. This investigation shows that the swelling of the cells occurs even if the mitotic activity is completely suppressed by the x-ray shock. It cannot be decided whether the increase in cell size is caused directly by the colchicine, or if this substance acts through the growth hormones present in the plant. In the former case, a new feature may be added to the collected picture of colchicine as a phytohormone given by HAVAS already in 1938.

Svalöf, Cyto-genetic Laboratory, November 1941.

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ALBERT LEVAN: Plant breeding by induction of polyploidy and some results in clover.

After the colchicine method had been introduced into plant breeding, whereby the production of polyploid plants was made an easy and a commonplace affair, it was often assumed that polyploid forms of great practical importance would soon appear. As new data in this field began to accumulate it was realized, however, that polyploidy *per se* is not a magic wand by means of which great practical results can be obtained. In spite of the fact that by now chromosome-doubled forms of most of the important crop plants have been produced, in no case has a polyploid variety proved to be immediately superior to its original form. The mere production of new polyploids must therefore be regarded only as the first step in a laborious breeding work.

We have become familiar, however, with certain factors, which determine the possibilities and the limitations of this kind of breeding work. *Firstly*, each plant species or even each genotype has an optimal range of chromosome numbers, within which its development is maximal. Above and below this range the viability is lowered, and at both ends the curve sinks into lethality. Unfortunately for plant breeders, most plants have presumably already during their evolution gravitated into their optimal range and therefore will respond with a decrease in viability to any change in their chromosome number. It must be remembered, however, that the environmental laws valid for cultivated plants are different from those in nature. *Secondly*, all autopolyploids have certain unfavourable properties in common. Their lower fertility is notorious and makes the breeding work in autopolyploid cereals, oil plants and other plants grown for seed production very difficult and very uncertain. Polyploids are also characterized by a slower growth, caused by their slower mitotic rhythm. It is known, however, that doubled varieties may, in spite of this, grow past their original forms and give a greater yield at the end of the season (compare, for instance, SCHLÖSSER, 1940).

The above considerations indicate that the prospects of breeding work in new autopolyploids should be more promising in plants, which in their spontaneous form have a low chromosome number, and the economical value of which lies in a great vegetative development, thus enabling the utilization of the much talked-of giant growth of the polyploids. That is the situation in the three *Trifolium* species, *Tr. pratense* (2x), *hybridum* (2x) and *repens* (4x). Wild *Trifolium* species with as high as 14-ploid and 20-ploid chromosome numbers are known and have a very good viability, so there is a good chance that the diploid and tetraploid species have not yet reached their range of optimal chromosome numbers. Chromosome-doubled forms of several commercial varieties of each of the three above-mentioned *Trifolium* species have been produced at our laboratory (LEVAN, 1940), and the first field trials with red clover and alsike clover were made last summer. Although the experiments were on a small scale and rather preliminary, they all gave

corresponding results and deserve a brief mentioning. A detailed account of our works on *Trifolium* will be published later.

Three populations of red clover and one of alsike clover were tested. Of each type one diploid and one tetraploid row, each with 30 individuals, were planted in the field. Further, 15 diploid and 15 tetraploid plants of a red clover clone, originated from cuttings from a 2x—4x chimaerical plant, were tested. Throughout these experiments the tetraploids yielded a considerably greater amount of green matter. The increase calculated on the weight of the diploids was 128 %, 36 %, 40 % in the red clover populations, 35 % in the clone and 31 % in the alsike. The dry matter content was somewhat lower in the tetraploids (the decrease = 2,1 % on an average). The quantity of crude protein in the dry substance of the leaves exhibited very small differences (tetraploid minus diploid = — 0,3, + 0,1, + 0,6 % in the population red clover, — 1,4 in the clone and — 0,5 in the alsike, the absolute values being 29,8—30,1, 31,1—31,0, 30,7—29,9, 25,9—27,3 and 29,3—29,8 %).

In my opinion these results show that *Trifolium* undoubtedly belongs to those plants from which one may be justified in expecting practical results from chromosome doublings. Much work still remains to be done before anything definite can be said about the practical value of the clover polyploids. The decrease in seed setting may turn out to be a crucial difficulty. Artificial pollinations and insect pollinations in cages show, however, that the inferior yield of seed obtained in the field during the last two years was at least partly due to a scarcity of insects.

Svalöf, Cyto-genetic Laboratory, December 1941.

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ALBERT LEVAN: The response of some flax strains to tetraploidy.

Flax belongs to those plants in which tetraploids are easily produced by the colchicine technique. Thus, as early as 1938 three instances of colchicine-induced flax tetraploids were published (GYÖRFFY, 1938; RYBIN, 1938; SIMONET, CHOPINET and SOUILIJAERT, 1938), and in 1939 MÜNTZING and RUNQUIST obtained tetraploid plants of a variety of oil flax. LUTKOV (1939) and LEVAN (1940) reported the production on a large scale of flax tetraploids. In the latter case about 400 c_1 tetraploids of 8 commercial varieties were secured (c_1 = the first generation after the colchicine-treated plants). Also tetraploids of another flax species, *Linum grandiflorum*, have been produced (SIMONET and CHOPINET, 1939).

So far, however, very little is known about the morphological qualities of tetraploid flax as compared with the diploids, beyond the fact that the tetraploids are distinguished by certain giant characters. In the present paper

some data will be given from certain of our tetraploid flax material, c_2 and c_3 of which have been grown in the field during the last two summers. The following 5 types have been studied closely: two typical fibre strains (Herkules, 041), one fibre strain characterized by certain resemblances to oil flax (Concurrent), and two typical oil strains (Szekacs, Palermo).

The tetraploids have certain properties in common as compared with the diploids. Thus, the general viability as well as the fertility is lower in the tetraploids throughout all experiments. This can easily be seen from a mere inspection of the field plots. A detailed study of certain properties, however, gives a more exact picture of the situation. In one experiment in 1940 the following mean values were obtained from 22 diploid and 22 tetraploid plants of each of the above-mentioned strains (the diploid—the tetraploid value is given for each strain, the same sequence of the strains as above being maintained). Number of side-branches: 5,3—2,0, 3,8—0,8, 7,6—1,0, 14,2—3,4, 8,0—2,5;

TABLE 1. *Relative values of the tetraploids (diploid values = 100).*

Studied property	Herkules	041	Concurrent	Szekacs	Palermo	Mean
Number of side-branches	37,7	21,1	13,2	23,9	31,3	25,4
Length of the longest side-branch	66,1	58,9	18,6	61,4	11,7	43,3
Flowering date	100,3	102,8	109,2	93,9	112,5	103,7
Number of seed capsules	53,6	25,5	35,8	58,6	40,7	42,8
Number of seeds per capsule	29,4	29,2	40,0	31,9	26,1	31,3
Dry matter weight	54,1	48,5	47,7	74,2	41,8	53,3
Petal length	116,2	120,2	112,7	115,9	105,6	114,1
1000-grain weight of the seeds	137,1	—	169,3	—	130,2	145,5
Plant height at the harvest	92,9	93,3	93,1	103,7	104,8	97,6

the length in cm. of the longest side-branch on July the 16th: 10,9—7,2, 5,6—3,3, 4,3—0,8, 10,1—6,2, 7,7—0,9; the first flower opened on the following dates: $^{6}/_{8}$—$^{6}/_{8}$, $^{22}/_{7}$—$^{24}/_{7}$, $^{25}/_{7}$—$^{30}/_{7}$, $^{23}/_{7}$—$^{29}/_{7}$, $^{2}/_{8}$—$^{30}/_{7}$; number of flowers forming seed capsules: 179—96, 157—40, 207—74, 169—99, 194—79; number of seeds per capsule: 9,2—2,7, 8,9—2,6, 9,0—3,6, 6,9—2,2, 6,9—1,8; weight in gm. of dry matter per plant: 36,2—19,6, 23,3—11,3, 35,4—16,9, 29,1—21,6, 29,2—12,2.

Besides a larger cell size the following giant characters of the tetraploids were observed: length in mm. of the petals: 11,7—13,6, 10,9—13,1, 11,8—13,3, 13,2—15,3, 12,6—13,3; the 1000 grain weight in gm. of the seeds: 4,67—6,40 (Herkules), 3,88—6,57 (Concurrent), 6,10—7,94 (Palermo). Tetraploid seeds of flax are rather characteristic in appearance, their colouring is mottled, which makes it possible to identify the tetraploids merely from the seeds.

The tetraploid strains behaved somewhat differently in one respect, viz. the course of the growth curve of the plant heights. All the plants of the above experiments were measured several times during their development. The tetraploids of the fibre flax were all the time on an average lower than the diploids, both the diploid and the tetraploid growth curve flattening out

towards the end of the season. In the oil flax, on the other hand, the tetraploids were at first lower than the diploids, but at about September the first the tetraploid curve crossed the diploid curve. At the harvest the following mean heights in cm. were obtained: 140—130, 124—116, 120—111, 84—88, 73—75. Evidently the tetraploid oil flaxes go on growing for a longer period than the tetraploid fibre strains.

The results are summarized in Table 1, where the mean values of the tetraploids are presented as relative numbers, corresponding diploid values being made equal to 100. As seen from the table, most of the important viability and fertility values of the tetraploids are considerably lower than 100. The comparison between tetraploid and diploid flax is rather discouraging from the plant breeder's view-point. There is little doubt that the tetraploids have passed the optimal range of chromosome numbers of the species.

It should be mentioned, however, that some evidence has come forward that tetraploid hybrids have a somewhat higher fertility than the pure tetraploid strains. Thus, 95 F_1 plants grown in the greenhouse yielded 17,3 seeds per plant, while 22 control tetraploids under the same conditions yielded only 8,9 seeds per plant. The increase is probably due to an improved general viability of the hybrids rather than a changed meiotic situation, as the number of seeds per capsule was the same in the hybrids as in the pure strains. A great many F_2 progenies were grown in the field last summer, showing considerable variations in plant viability. A fuller account of these experiments on tetraploid flax is under preparation.

Svalöf, Cyto-genetic Laboratory, December 1941.

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